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CONTENTS

Sawfly resistance in wheat. IV. Some effects of light intensity on resistance — *D. W. A. Roberts and C. Tyrell* 457
Progeny performance of seven red raspberry varieties in Nova Scotia — *L. E. Aalders and D. L. Craig* 466
Comparison of tissues from solid- and hollow-stemmed spring wheats during growth. I. Dry matter and nitrogen contents of pith and wall and their relation to sawfly resistance — *A. J. McGinnis and R. Kasting* 469
Crop rotations and common rootrot in wheat — *R. J. Ledingham* 479
Variability within isolates of *Verticillium albo-atrum* from potato — *D. B. Robinson* 487
Inherent differences in depth of crown in wheat and barley — *B. J. Sallans* 493
Variation in the sensitivity of isolates of *Venturia inaequalis* (CKE.) Wint. to fungicides — *R. G. Ross and S. A. Hamlin* 499
Petal blight, a new disease of the canna "Pfitzer's Dwarf" — *H. S. Thompson* 503
The organic acids in silage as determined by gas chromatography — *J. R. Lessard, R. A. Briggs and J. V. Scaletti* 507
Response of little cherry-infected cherry trees to zinc treatments — *J. M. Wilks and J. A. Stewart* 517
Flower, pod and seed development relative to the timing of the seed harvest of Viking birdsfoot trefoil (*Lotus corniculatus*) — *J. E. Winch and H. A. Macdonald* 523
Performance of four irrigated pasture mixtures under grazing by sheep — *D. B. Wilson and R. D. Clark* 533
Host range studies of the little cherry disease virus — *Jack M. Wilks and Maurice F. Welsh* 544
Row width and seeding rate in relation to seed production in timothy, *Phleum pratense* L. — *R. S. Fulkerson and W. E. Tossell* 549
Honeybee activity on safflower (*Carthamus tinctorius* L.) — *R. Boch* 559
Evapotranspiration at Kapuskasing, Ontario — *L. J. Chapman and P. Dermine* 563
Studies on the control of dwarf bunt in winter wheat — *S. G. Fushtey* 568
Some effects of four chlorinated polycyclic insect toxicants on the physiology of potatoes, carrots and radish — *A. M. Unrau and G. Howell Harris* 578
The inheritance of rust resistance. VII. The inheritance of resistance to races 15B and 56 of stem rust in eleven common wheat varieties of diverse origin — *D. R. Knott and I-sun Shen* 587
Comparison of grasses for dryland turf — *R. P. Knowles* 602
Inheritance of quality as related to agronomic characters in advanced lines of a spring wheat cross — *F. Gfeller and A. G. O. Whiteside* 607
Comparison of lightly grazed and ungrazed range in the fescue grassland of southwestern Alberta — *Alexander Johnston* 615
Induction of seed set in dwarf-spring and winter wheat — *E. A. Hurd* 623
Effects of clipping and nitrogen on competition between three pasture species — *D. B. Wilson and W. S. McGuire* 631
Phenology of several plant species at Ottawa, Ontario, and an examination of the influence of air temperatures — *I. J. Bassett, R. M. Holmes and K. H. MacKay* 643
An evaluation of varietal characteristics of orchardgrass (*Dactylis glomerata* L.) subjected to a silage-pasture type of utilization — *W. E. P. Davis* 653
The persistence of certain soil insecticides for control of the tuber flea beetle, *Epitrix tuberis* (Gent.), in the interior of British Columbia — *F. L. Banham* 664
Notes on:
Bismaleamic acid as a tobacco sucker growth inhibitor — *E. V. Parups and I. Hoffman* 672
The growth characteristics of alfalfa and red clover plants derived from hard seeds and seeds of different sizes — *Dale Smith* 675
A greenhouse technique for crossing legumes by honey bees — *B. P. Goplen and P. Pankiw* 679
Russell oats — *F. J. Zillinsky* 683
Wild oat control by delayed seeding of wheat — *N. A. Korven* 685
Holder-lifter tongs for harvesting forage rows — *H. Sivers and B. E. Twamley* 687
Chemical changes occurring in freeze-dried and fresh-frozen wheat leaves during storage — *Harold J. Perkins* 689

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SAWFLY RESISTANCE IN WHEAT

IV. SOME EFFECTS OF LIGHT INTENSITY ON RESISTANCE[1]

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[Received for publication September 21, 1960]

ABSTRACT

The degree of resistance to the wheat stem sawfly (*Cephus cinctus* Nort.) of seven varieties of wheat grown under shaded conditions was shown to be lower than that of plants grown under unshaded conditions. The breakdown of resistance of Rescue wheat grown in the greenhouse in the winter was prevented by a high-intensity light supplement of 4000 foot-candles but not by a supplement of 1500 foot-candles. The low resistance of Rescue wheat grown in the greenhouse results from low light intensities in the greenhouse in both summer and winter. It was concluded that high light intensities are required for the maximum expression of stem-solidness and sawfly resistance in Rescue wheat.

INTRODUCTION

It has been shown (7) that the resistance to wheat stem sawfly (*Cephus cinctus* Nort.) of seven varieties of wheat tested was lower when the plants were grown in the greenhouse than when they were grown outdoors. Subsequent work (8) showed that, regardless of the stage of development of the plants, Rescue wheat grown in the greenhouse during the winter months had little resistance to the development of eggs and establishment of first-instar larvae.

Since the soil used for the summer greenhouse experiments and their outdoor controls was from the same source (7), it is unlikely that the differences in resistance were caused by differences in mineral nutrition of the plants. It was pointed out earlier (6) that temperature is not likely the major factor causing the breakdown of resistance of plants grown in the greenhouse but that light intensity might be. Measurements with a Weston illumination meter, Model 756, showed that light intensity in the greenhouse was only about 75 per cent of that outdoors. Platt (4) observed that shading reduced the stem solidness of wheat. Later, Platt and Farstad (5) found that the resistance of S-615 grown under cages was lower than that of plants grown in the open and that there was a close association between a small number of hours of June sunshine and a high percentage of stems cut by sawfly in S-615. Working with plots shaded with muslin, Roemhild (9) found that the resistance of Rescue and Golden Ball wheat was lower when the plants were grown in the shade.

These results suggest that low light intensity or unsuitable light quality may be the cause of the low resistance of Rescue wheat grown in the

[1]Contribution from the Plant Pathology Section.

greenhouse. This paper presents the results of experiments designed to test this hypothesis. Two types of test were made, as follows: first, experiments on shading comparable to those of Roemhild and, second, experiments with light supplements in the greenhouse in the winter.

MATERIALS AND METHODS

Experiments with Shading

The same seven varieties of wheat were used as in the previous tests (7), namely, the bread wheats Thatcher, Red Bobs, Rescue, Hybrid H4191, and Hybrid H46146, and the durum wheats Golden Ball and Melanopus.

The shading experiments were replicated twice inside a large bronze-screened cage located on irrigated land close to the control plots. The light intensity inside this cage was 50 to 60 per cent of the full outdoor intensity as determined by a Weston illumination meter, Model 756. These plots were seeded at the same time as the control plots described earlier as the outdoor plot tests (7). The shaded plots were infested by sawflies liberated in the cage when the wheat was between shot-blade and flowering.

When the wheat was ripe and the cutting of the stems by sawflies was complete, the material was dug up and counts were made as described earlier (6, 7). Stem solidness was determined by a modification (7) of the method of Larson (2).

The data on sawfly resistance and stem solidness were subjected to the analysis of variance. Where percentages were involved, the arcsin transformation was used. The data were combined for analysis as indicated in the tables and whenever a significant interaction was found it was used to test the significance of the treatment or varietal differences.

Experiments with Light Supplements

The reaction of Rescue wheat in the greenhouse in the four winters involved was used as a control in the experiments with various light supplements. Some of the early experiments with low-intensity light supplements were based on the assumption that the filtering action of the glass in the greenhouse was altering the quality of light incident on the plants. These supplements failed to increase sawfly resistance and so a high light intensity incandescent light bank capable of supplying plants with 3000- to 4000 foot-candles was constructed. This bank consumed approximately 13 kilowatts of electrical power.

In all of the experiments with low-intensity light supplements the additional light was supplied continuously. For some tests two 40-watt fluorescent tubes which produced a supplement of approximately 200 foot-candles were used. Other tests were made with one 300-watt incandescent neck-reflector bulb, supplying an extra 800 foot-candles of light to the plants. Two other trials were made with three 300-watt neck-reflector incandescent light bulbs. The plants in these tests were cooled with one 10-inch desk fan. The additional light at the plant level was 1500- to 1800 foot-candles. Although only two trials with some of these supplements were used, the negative results are presented because they help to define the type of supplement that is required to produce an effect.

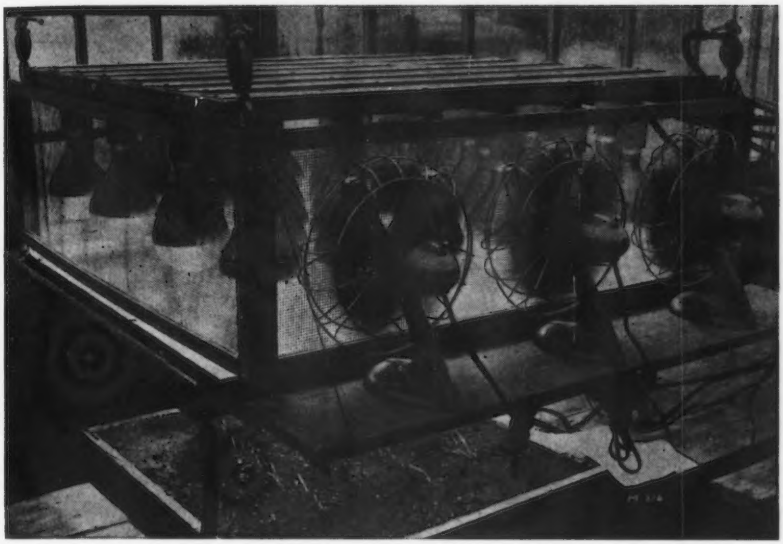

FIGURE 1. The high-intensity light bank producing 3000 to 4000 foot-candles at a distance of 100 centimetres.

The high-intensity light bank (Figure 1) had an angle-iron frame with outside dimensions of 42 by 39½ by 16½ inches. Mounted on top of the frame were seven galvanized iron channels, spaced 5¼ inches apart. These were 1¼ inches deep and 1½ inches wide, with additional half-inch wide flanges on each side. Each channel was covered with a sheet of galvanized iron. Each channel carried the wiring for six 300-watt neck-reflector incandescent light bulbs (Canadian General Electric C 300 RH Mogul base), which fitted into porcelain sockets mounted at 6-inch centres. The sockets were staggered on adjacent channels to space the bulbs more closely. The bottom of the angle-iron frame was closed with a piece of plate glass sealed with a suitable grade of Tremco caulking compound. On three sides of the glass there was a 1½-inch-high watertight barrier, but on the fourth side there was only a ridge ½-inch high, which acted as an overflow to control the depth of cold water on top of the plate glass. On the outside of this ridge there was a watertight trough of sheet metal connected to a drain. A layer of cold water at least ½-inch deep over the glass was maintained by continuously supplying water through a pipe with holes at intervals.

Removable wire-mesh screens were mounted on all sides of the angle-iron frame to prevent accidents from flying glass if a bulb exploded. The light bulbs were cooled by three 10-inch desk fans mounted on a wooden platform supported by two angle-iron brackets. The whole unit was suspended by four cords passing over pulleys and so could be raised or lowered to adjust the light intensity at the plant level.

TABLE 1. — SOLIDNESS RATINGS AND PERCENTAGES (TRANSFORMED VALUES[1]) OF STEMS IN WHICH THE WHEAT STEM SAWFLY DIED AT VARIOUS STAGES FOR VARIOUS WHEAT VARIETIES GROWN WITH AND WITHOUT SHADE

Wheat variety	Solidness rating[2]		Percentage of infested stems not cut		Percentage of infested stems with eggs that died		Percentage of stems with tunnelling larvae that died	
	Not shaded	Shaded	Not shaded	Shaded	Not shaded	Shaded	Not shaded	Shaded
Bread wheats								
Hollow-stemmed								
Thatcher	8	8	67	45	18	13	65	42
Red Bobs	7	7	70	41**	9	13	70	40**
Solid-stemmed								
Rescue	26	18**	71	45**	30	11**	68	43**
H4191	27	21*	73	37**	31	12**	70	34**
H46146	25	21	73	44**	34	15*	70	40**
Durum wheats								
Melanopus	23	21	64	51	14	18	62	46
Golden Ball	33	32	71	58	16	34*	70	47
L.S.D.[3]	3	2	—	—	7	6	—	—

[1]Arcsin transformation (10). Averages of duplicate tests run for 3 years
[2]Modification (7) of method of Larson (2). High values represent greater solidness
[3]Where no value is given, differences between varieties were not significant
*Values for shaded plants significantly different from those for unshaded plants at 5% level
**Values for shaded plants significantly different from those for unshaded plants at 1% level

In the experiments with this high-intensity light bank plants were grown in soil bins similar to those used for the winter greenhouse tests. In the earliest experiments the soil on the surface of the bins became overheated and killed the plant tissue at the soil surface (1). This was overcome by using three extra 10-inch desk fans to cool the soil surface. The intensity of the light supplement was approximately 3000-4000 foot-candles until the plants reached shot blade, when it was reduced to approximately 2000 foot-candles at the level of the ears.

Plants grew to maturity satisfactorily under continuous illumination from the high-intensity light bank. They were shorter and more wiry than those grown in the greenhouse in the winter and looked like plants grown outdoors in the summer on a dry windy knoll.

Only two experiments with Rescue wheat were made with this light bank. Except as stated above the plants and sawflies were handled in the same way as those in the winter greenhouse experiments (7). These experiments should be repeated with Rescue wheat and also other varieties but the senior author is no longer studying sawfly resistance in wheat.

RESULTS

Experiments with Shading

The results of 3 years of experimentation are summarized in Table 1. The data show a trend toward lower stem solidness in the solid-stemmed bread wheats grown under shaded conditions. The difference was significant only for Rescue and Hybrid H4191.

The percentage of infested stems not cut was lower under shaded conditions than under full sunlight. This difference was highly significant in Red Bobs and the solid-stemmed bread wheats. An examination of the data on the percentage of infested stems with eggs that did not hatch and of stems with tunnelling larvae that did not survive shows that both these types of resistance contributed to the higher level of total resistance in the solid-stemmed bread wheats grown under unshaded conditions. In Thatcher, Red Bobs, and Melanopus shading produced no significant difference in the percentage of infested stems with eggs that did not hatch, whereas in Golden Ball shading produced a significant increase in the percentage of infested stems with eggs that did not hatch. In all cases the value for the percentage of stems that were tunnelled but not cut was higher under full sunlight than under shaded conditions. This difference was highly significant in the solid-stemmed bread wheats and Red Bobs but not in the other varieties.

Experiments with Light Supplements

Data on the resistance of Rescue wheat grown in the greenhouse in winter are based on experiments in which 39 to 134 stems were infested in each replication. Because the distribution of values obtained from the replications was apparently not normal, frequency distributions are substituted for the mean values and standard errors. The percentage of stems

infested but not cut was low in most cases and the values for the 16 replications were distributed as follows:

0–10%	10–20%	20–30%	30–40%	40–50%	50–60%	60–70%
4	4	2	4	1	0	1

With one exception the percentage of stems with eggs that did not hatch was low and the values were distributed as follows:

0–5%	5–10%	10–15%	15–35%	35–40%
10	4	1	0	1

The percentage of stems that were tunnelled but not cut was also low with the following distribution of values:

0–10%	10–20%	20–30%	30–40%
4	1	6	5

Except for the 4000 foot-candle incandescent light supplement the light supplements used produced either no, or only little, change in resistance in all varieties (Table 2). Apparently a light supplement in excess of continuous illumination with 1500 foot-candles of incandescent light is required to modify the sawfly resistance of Rescue wheat appreciably. With a continuous light supplement of 4000 foot-candles there was an appreciable increase in percentage of infested stems with eggs that failed to hatch and also in percentage of tunnelled stems not cut. These changes in resistance are opposite to those produced in Rescue wheat when grown under shaded conditions.

The data in Table 3 show that only the 4000 foot-candle incandescent light supplement affected stem solidness to any extent. These data support the conclusions of earlier workers (4, 5) that low light intensities reduce stem solidness and that a high threshold of insolation is needed for the full expression of stem solidness in Rescue.

DISCUSSION

The data presented show that high light-intensities favour 1) the development of a high degree of stem solidness in the solid-stemmed bread wheats, 2) a high level of resistance to the development of eggs and young first-instar larvae of the wheat stem sawfly in solid-stemmed bread wheats, and 3) a high level of resistance to the development of older larvae in

TABLE 2. — PERCENTAGES OF INFESTED STEMS IN WHICH THE WHEAT STEM SAWFLY DIED AT VARIOUS STAGES IN RESCUE WHEAT GROWN IN THE GREENHOUSE IN THE WINTER WITH VARIOUS TYPES OF CONTINUOUS SUPPLEMENTARY LIGHT

Type of supplement	Number of stems infested	Intensity, foot-candles	Per cent infested stems with eggs that did not hatch	Per cent tunnelled stems not cut	Per cent infested stems not cut
Fluorescent, 40-watt					
2 4500° K-white tubes	64	200	6	15	20
2 4500° K-white tubes	37	200	5	17	22
2 4500° K-white tubes	66	200	0	41	42
1 blue and 1 4500° K-white tube	63	—	0	17	17
1 blue and 1 4500° K-white tube	32	—	0	28	28
1 black light and 1 4500° K-white tube	33	—	0	36	36
1 black light and 1 4500° K-white tube	38	—	0	5	5
Incandescent, 30-watt					
1 bulb	39	800	0	38	38
1 bulb	92	800	0	12	12
3 bulbs	47	1500	15	32	43
3 bulbs	41	1500	2	22	24
Bank of 42 bulbs	58	4000	27	93	95
Bank of 42 bulbs	24	4000	33	62	75

TABLE 3. — STEM SOLIDNESS RATINGS OF RESCUE WHEAT GROWN IN THE GREENHOUSE IN WINTER WITH VARIOUS TYPES OF CONTINUOUS SUPPLEMENTARY LIGHT

Type of supplement	Solidness rating
None	21
Fluorescent, 40-watt	
2 4500° K-white tubes	19
1 blue and 1 4500° K-white tube	21
1 black light and 1 4500° K-white tube	21
Incandescent, 300-watt	
1 bulb	20
3 bulbs	20
Bank of 42 bulbs	35

solid-stemmed bread wheats. It is apparent that hours of sunlight and light intensities of the order of those received in the Lethbridge area were required for the full expression of resistance to the sawfly in solid-stemmed bread wheats. Breakdown of resistance in the greenhouse was apparently caused by the low light intensity. The failure of short-wave length light supplements (blue or black light fluorescent) to increase resistance and stem-solidness suggests that the differential filtering action of greenhouse glass, which greatly reduces the light intensity in the ultra-violet region, was not responsible for the breakdown of resistance in the greenhouse.

This view is supported by the success of the incandescent light bank in promoting stem-solidness and resistance. Incandescent lights are poor sources of ultra-violet and in our experiments this light was filtered through plate glass.

These results offer a possible explanation of the results obtained in the 1953 experiments with different dates of seeding. The data (8) show that, if the results of infestations at comparable stages of plant development are considered, the resistance of the plants seeded June 16 was much greater than that of those seeded May 13 or May 27. In Thatcher, Red Bobs, and Rescue the resistance, as measured by the percentage of stems tunnelled but not cut, in the plants seeded June 16 was greater than that in those seeded May 13 and 27 if we consider exposure to infestation at the same stage of development of the plants. In Rescue the percentage of infested stems with oviposition scars was higher in the Week 4 infestation of plants seeded June 16 than the Week 2 infestation of plants seeded May 13 or 27. That the total solar radiation received during the early stages of development of the plants seeded June 16 was considerably higher than that received by the plants seeded earlier is shown by the following measurements made by the Meteorological Division, Canada Department of Transport, at a location approximately 15 miles from the plot area (3):

Period	Total radiation gm.-cal. per sq. cm.*	
May 13-27	7580	First date of seeding, May 13
May 28-June 10	6733	Second date of seeding, May 27
June 11-24	9614	Third date of seeding, June 16
June 25-July 8	9157	
July 9-22	10198	First exposure to sawflies, July 6
July 23-Aug. 5	8665	Second exposure to sawflies, July 20
Aug. 6-19	8271	Third exposure to sawflies, Aug. 3
Aug. 20-Sept. 2	7445	Fourth exposure to sawflies, Aug. 17

*The meteorologists state that these figures are probably too high

This suggests that the critical stage for the production of a high degree of sawfly resistance occurs early in the development of the plant. This is in agreement with the conclusions of Platt and Farstad (5), that long hours of June sunshine induce a high level of sawfly resistance in potentially resistant wheat varieties.

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PROGENY PERFORMANCE OF SEVEN RED RASPBERRY VARIETIES IN NOVA SCOTIA[1]

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[Received for publication January 25, 1961]

ABSTRACT

Progeny from 21 crosses among 7 red raspberry varieties were evaluated with respect to yield, winter hardiness and resistance of canes to anthracnose (*Elsinoe veneta*). Seedlings of the varieties Trent and Rideau showed the greatest bud survival, especially when crossed with other varieties. The highest yielding progeny was Malling Promise x Trent and this was not changed by adjusting yields for bud survival or number of canes per plot. Resistance to anthracnose was found in progenies of Viking, Willamette and Early Red and inheritance of anthracnose resistance seemed to be additive.

INTRODUCTION

Red raspberry clones vary in their ability to produce desirable progeny. As early as 1913, Wellington (5) advised workers to "test the prepotency of varieties before using them in breeding". Reports published since then (1, 2, 3, 4) indicate that the European varieties Lloyd George and St. Walfried give the most desirable progeny, especially when crossed with such American varieties as Viking, Newburgh and Cuthbert. However, these statements are generally based on field observations of seedlings grown in widely scattered geographic areas in non-replicated trials and for which direct comparisons on a statistical basis are impossible. The work reported here was conducted, therefore, to evaluate, under Nova Scotia conditions, those parents that were said to give desirable progeny elsewhere, and to compare them directly with others whose progeny performance was relatively unknown.

MATERIALS AND METHODS

The seven varieties of red raspberries used were: Early Red (Lloyd George x Cuthbert), Lloyd George (chance seedling), Malling Promise (Newburgh x (Lloyd George x Pynes Royal)), Rideau (Lloyd George x Newman 23), Trent (Newman 23 x Lloyd George), Viking (Cuthbert x Marlboro), and Willamette (Newburgh x Lloyd George). The seven varieties were crossed in all combinations, omitting reciprocals. The seeds were germinated in the greenhouse, and sufficient seedlings were obtained to permit the selection of reasonably uniform progenies for field planting. Plots of eight seedlings from each progeny were set in a randomized complete block, replicated four times. The rows were planted 10 feet apart and the seedlings 2 feet apart in the row with 5-foot gaps at the ends of plots. The plots were clean cultivated throughout the growing season and, where possible, were pruned uniformly to five canes per plant each April. Records on winter injury, yield, and incidence of anthracnose were taken during 1959 and 1960. Winter injury was determined by counting the total number of buds on each cane, the number successfully breaking in the spring, and calculating the per cent of buds breaking for each plot. Yields

[1]Contribution No. 1059 from the Research Station, Canada Department of Agriculture, Kentville, N.S.

were taken in grams per plot with pickings being made every second or third day. Incidence of anthracnose was scored following examination of the canes in October. The bud survival and anthracnose data, expressed as percentages, were transformed using the angular transformation prior to performing a variance analysis.

RESULTS AND DISCUSSION

An initial analysis of variance showed highly significant differences among plots for all three characters studied. Significant differences between specific progenies were then determined, using Duncan's Multiple range test.

Progenies having Trent and Rideau as parents were generally quite winter-hardy. The seven progenies showing the greatest bud survival had one or the other of these varieties as one parent, but the cross between the two ranked only tenth and was significantly poorer than the top seven crosses (Table 1). The reason for this is not apparent. Progeny of the

TABLE 1. — PERFORMANCE OF 21 RED RASPBERRY PROGENIES AS MEASURED BY BUD SURVIVAL, YIELD, AND INCIDENCE OF ANTHRACNOSE

Parentage	Bud survival		Yield		Incidence of anthracnose	
	Mean	Significance[1]	Mean	Significance	Mean	Significance
	(%)	(5%)	(gm.)	(5%)	(%)	(5%)
Early Red x Lloyd George	28.8	hi	827	defg	62.6	cdef
x Malling Promise	30.7	h	1067	bcdef	38.1	ab
x Rideau	46.8	b	1028	bcdef	65.0	cdef
x Trent	44.9	bc	1201	bcde	50.1	bcd
x Viking	38.3	f	1085	bcdef	27.5	a
x Willamette	34.6	g	1173	bcde	30.6	a
Lloyd George x Malling Promise	33.3	g	1280	bc	78.1	f
x Rideau	32.9	g	567	g	68.9	def
x Trent	51.2	a	1313	abc	76.2	f
x Viking	33.6	g	1065	bcdef	67.2	def
x Willamette	39.2	f	1065	bcdef	50.6	bcd
Malling Promise x Rideau	34.9	g	1068	bcdef	65.0	cdef
x Trent	44.2	cd	1719	a	67.5	def
x Viking	29.0	hi	874	cdefg	49.4	bc
x Willamette	35.0	g	1320	abc	38.8	ab
Rideau x Trent	37.4	f	1020	bcdef	65.0	cdef
x Viking	42.8	de	822	defg	73.8	ef
x Willamette	42.1	e	1340	ab	65.6	cdef
Trent x Viking	37.2	f	714	fg	68.1	def
x Willamette	41.7	e	1252	bcd	58.1	cde
Viking x Willamette	27.2	i	778	efg	24.9	a

[1] Any two means not followed by the same letter are significantly different

cross Lloyd George x Trent had significantly greater bud survival than all other progenies.

The highest yielding progeny was Malling Promise x Trent and it was significantly better than all others except three. The lowest yielding progeny was Lloyd George x Rideau. Some of the differences in yield were no doubt due to winter injury to the buds and to the fact that some plants produced less than five canes. To correct for this, a regression analysis for yield on number of canes per plot and on per cent surviving buds was calculated. A significant reduction in sums of squares resulted with the partial regression coefficients for bud survival and canes per plot being +12.1 and +28.3, respectively. When the yields were adjusted for both these factors Malling Promise x Trent was still the highest yielding progeny and Lloyd George x Rideau and Trent x Viking were still the two lowest yielding progenies. Even though the number of canes and per cent surviving buds affected the yields significantly, it would appear that this was a minor factor compared to genetic differences in the yielding abilities of the different progenies.

The three parents giving progeny with the least amount of anthracnose were Viking, Willamette and Early Red. The best three progenies from the standpoint of incidence of anthracnose were the three combinations of these parents. One or another of them was involved as one parent in each of the next best seven progenies. It would, therefore, appear that factors for resistance to anthracnose are additive in their action. Lloyd George seemed to confer the least resistance to anthracnose on its progeny, although the variety itself is not particularly susceptible to the disease. Again there appears to be little correlation between yield and incidence of anthracnose.

As a group, the seven varieties used in this study were very closely related genetically. If a wider range of varieties and a larger number of seedlings per progeny had been used, it is likely that a much greater divergence in progeny performance would have been found.

Even within these closely related types, the progeny test showed quite significant differences in combining abilities. It would, therefore, appear that it is well worthwhile to get some indication of the value of a parent through a progeny test before it is used extensively in a breeding program.

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COMPARISON OF TISSUES FROM SOLID- AND HOLLOW-STEMMED SPRING WHEATS DURING GROWTH

I. DRY MATTER AND NITROGEN CONTENTS OF PITH AND WALL AND THEIR RELATION TO SAWFLY RESISTANCE[1]

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ABSTRACT

The dry matter content of the internodes of the wheat varieties, Thatcher, Rescue, and Golden Ball, grown on non-irrigated land varied with age, variety, and internode. The nitrogen content of the stem tissues varied from 6 per cent for immature to less than 1 per cent for mature tissues. The nodes of these wheat plants generally contained less dry matter and more nitrogen than did the internodes. At maturity the top third of each of the two uppermost internodes of Thatcher grown on irrigated land contained the highest percentages of dry matter and nitrogen, and the bottom third contained the lowest. The stems of the wheats and of Eagle oats were not greatly different in dry matter and nitrogen content during the period when larvae of the wheat stem sawfly, *Cephus cinctus* Nort., usually die in oat plants.

Resistance of solid-stemmed wheats to the sawfly does not appear to be associated with a quantitative nutritional deficiency of either moisture or nitrogen. The analyses are consistent with the hypothesis that older larvae in solid-stemmed wheats die as a result of desiccation. Pith of Golden Ball contained more moisture than did that of Rescue, which supports the view that greater egg mortality may occur in the former variety because of the higher moisture content.

INTRODUCTION

Solid-stemmed wheats are generally resistant to the wheat stem sawfly, *Cephus cinctus* Nort., whereas hollow-stemmed varieties are susceptible (11). Oat plants, although hollow-stemmed, are immune to sawfly damage (2). If the piths of solid-stemmed wheats or the stem tissues of oats are nutritionally inadequate for larvae of the sawfly, the resistance might be explained. Further, the sawfly selects an immature portion of the growing stem for oviposition. Consequently, as an internode develops, the eggs are laid first in the top portion and then, as these tissues mature, progressively lower in the internode (5). Perhaps the oviposition sites have particular moisture and nitrogen contents that affect hatching and early larval development, respectively.

An earlier publication (8) showed that in solid-stemmed wheats greater differences in moisture and nitrogen content existed among the internodes of a stem than between the pith and wall tissues from the same internode. However, sawfly larvae feeding in plants of the age analysed (80 and 98 days after seeding) could have matured without further feeding. Thus it was of interest to determine the percentages of dry matter and nitrogen in pith and wall tissues from solid-stemmed wheats at earlier stages of growth.

This paper shows the dry matter and nitrogen content by internode of the pith and wall tissues of two solid-stemmed wheats and of the wall tissues of a hollow-stemmed wheat and of an oat variety from early shot-blade to maturity. Some implications of these data relative to resistance to the sawfly are considered.

[1]Contribution from the Entomology and Chemistry Sections.

MATERIALS AND METHODS

Three spring wheats, Thatcher and Rescue (*Triticum aestivum* L. emend Thell.) and Golden Ball (*T. durum* Desf.), and one oat variety, Eagle (*Avena sativa* L.), were grown on non-irrigated land at Lethbridge in randomized eight-row plots. Samples were taken during the growing season, beginning on June 20, when the plants were in the early shot-blade stage. The final samples of Thatcher, Rescue, and Eagle were taken on August 13. Golden Ball matures more slowly than the other varieties, and therefore an additional sample was taken on September 5. On each collection date three plots of each of the wheats and two of the oats were sampled and analysed separately. The percentages of dry matter and nitrogen for the wheats are mean values from the three plots.

Samples consisted of about 25 stems from the middle six rows of each plot. They were immediately taken to the laboratory and all leaves carefully removed from the stems. The stems were then divided into internodes, which were numbered from the crown and treated separately. The pith was stripped from the internodes of Rescue and Golden Ball. When they were immature it was not possible to separate the pith and wall tissues.

The nodes from each variety and collection date were combined for analyses of dry matter and nitrogen.

The dry matter and nitrogen contents of sections of internodes 4 and 5 of Thatcher stems grown on irrigated land were also determined. Samples were taken in triplicate at approximately weekly intervals beginning June 21, when the plants were at the early shot-blade stage. Although the plants were ripe on August 12, a final collection was made on September 6. Where possible, each internode was divided into three approximately equal segments. The third internode was well developed in plants collected on June 21, but the fourth and fifth internodes together were only 1 to 3 centimetres in length. For this sample the fourth and fifth internodes were neither separated nor divided. On the second date, only the fifth internode was short and it was not divided. The fourth internode was divided into meristem and mature tissues at that point where the internode broke cleanly when the ends were brought together. The mature portion was in turn subdivided into approximately equal segments, referred to as top and middle. All subsequent samples consisted of the fourth and fifth internodes, each of which was divided into three approximately equal segments, designated top, middle, and bottom.

The first sample of Eagle oats grown on irrigated land was collected on July 9. Each of the two uppermost internodes was divided into three segments, as was done for Thatcher. On subsequent collection dates a single sample of 10 to 15 representative stems was processed.

The dry matter content was determined on all samples by drying *in vacuo* at 90°C. for 8 hours (1). The dried samples were ground in a micro-Wiley mill to pass a 40-mesh screen and the nitrogen content of each was determined in duplicate by the micro-Kjeldahl method (1).

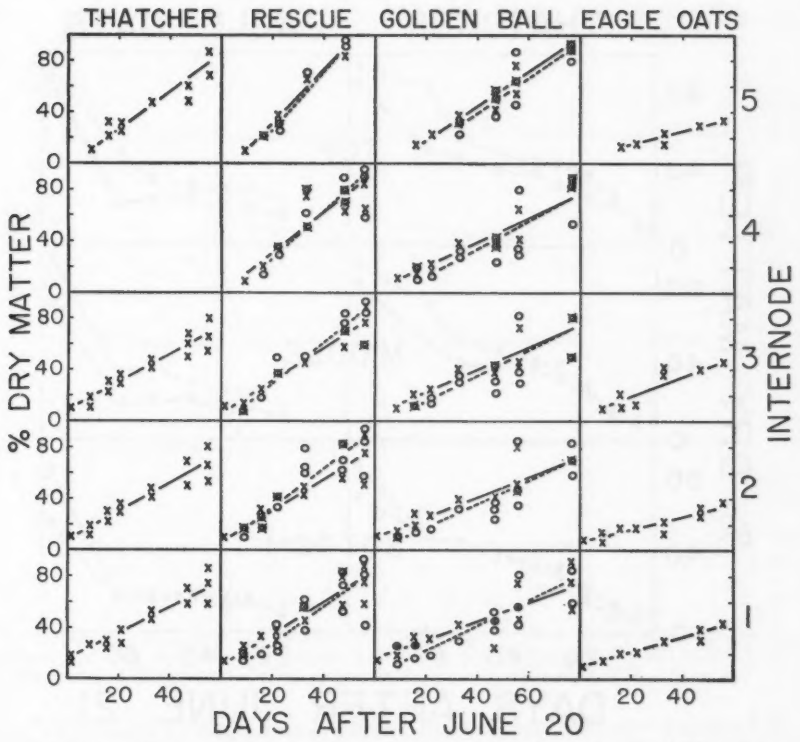

FIGURE 1. Dry matter contents of stem tissues from three wheat varieties and one oat variety grown on non-irrigated land. Internode 4 of Thatcher and Eagle was labelled internode 5. X—X wall; 0—0 pith.

RESULTS AND DISCUSSION

Composition of Tissues During Growth

The stems of Thatcher and Eagle grown on non-irrigated land had only four internodes, whereas the stems of Rescue and Golden Ball wheat had five. The top internodes were compared because they are structurally similar. The changes in the dry matter content* of the different internodes (Figure 1) quantitatively confirm field observations. As the plants matured the percentage of dry matter generally increased in all the stem tissues. The stems of Golden Ball matured more slowly than did those of either Thatcher or Rescue, as indicated by the slower rate of increase in dry matter. The slower development of Golden Ball is also shown in Figure 1 by the dates on which the various internodes of the three wheat varieties were first sampled. On the first date the bottom three internodes of both Thatcher and Rescue were collected whereas only the bottom two internodes from Golden Ball had differentiated sufficiently to permit collection. On the second date the full complement of internodes in Thatcher and

*Percentage dry matter = 100 minus percentage moisture

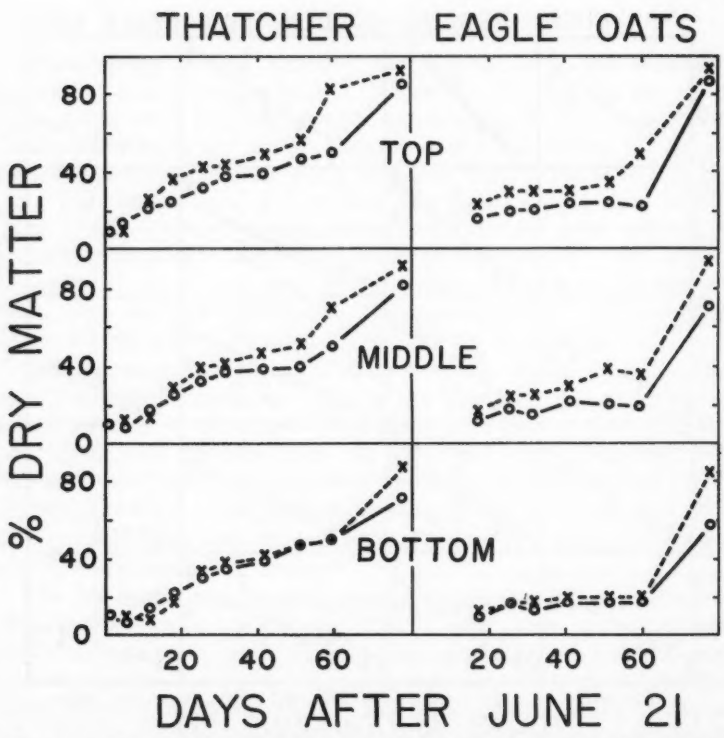

FIGURE 2. Dry matter contents of sections of internodes 4 and 5 from Thatcher wheat and Eagle oats grown on irrigated land. 0—0 internode 4; X---X internode 5.

Rescue were collected. At this time the fifth internode of Golden Ball had not differentiated and was first collected on the third date.

Regression coefficients of the changes in dry matter content with time, calculated for the data plotted, were significant ($P < 0.01$). The rates of drying of the internodes within a variety, represented by the plotted regression equations in Figure 1, were not different ($P > 0.05$). However, there was a tendency for the top internodes of the wheats to lose moisture most rapidly and for the second internode from the top to be intermediate in rate of drying between the top internode and the lower ones.

The rates of drying of the pith and wall tissues for either Rescue or Golden Ball, as measured by regression coefficients, were not different ($P > 0.05$). The differences in dry matter content between pith and wall were not statistically significant ($P > 0.05$). None the less, the consistent pattern for dry matter content of pith and wall for the internodes within a solid-stemmed variety throughout the growing period suggests that the differences are real. The way in which the dry matter content of the pith changed in relation to that of the wall during growth was different for the two solid-stemmed varieties.

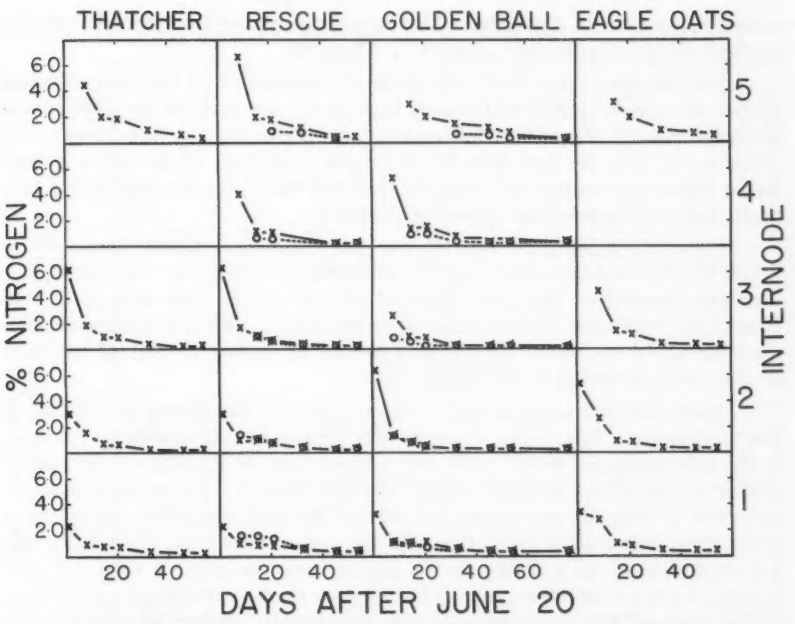

FIGURE 3. Percentage of nitrogen on dry weight basis in stem tissues from three wheat varieties and one oat variety grown on non-irrigated land. Internode 4 of Thatcher and Eagle was labelled internode 5. X—X wall; 0—0 pith.

The stems of Eagle oats grown on non-irrigated land also increased in dry matter content during the growing period (Figure 1). The most striking difference between the stems of wheat and those of oats was the slower rate of drying of the latter during growth. On the seventh sampling date the oat stems contained about 40 per cent dry matter whereas the stems of Thatcher and Rescue contained nearly 80 per cent and the dry matter content of Golden Ball stems was near 60 per cent. The dry matter content of all internodes of Eagle increased at about the same rate and attained the same level whereas in wheat stems it increased more rapidly, and reached the highest level in the two uppermost internodes.

Except for the last sampling date, the dry matter contents of the segments from internodes 4 and 5 of Thatcher were consistently higher than those of the corresponding parts of Eagle (Figure 2). There was a tendency for the segments of the fifth internode of both Thatcher and Eagle to be drier than comparable segments of the fourth internode. With the exception of one date the top third of internode 5 of Thatcher had a higher dry matter content than did any of the other portions of the two uppermost internodes. The fourth and fifth internodes dried from the top downward but smaller differences occurred in the former. The different segments of the top two internodes of Eagle showed only slight changes in dry matter content during growth until the end of the season, at which time the dry

matter content rose abruptly. This is in contrast to the generally steady increase in the dry matter content of Thatcher.

The nitrogen content of the different internodes of the four varieties grown on non-irrigated land was as high as 6.7 per cent of the dry weight during the early stages but did not exceed 2.2 per cent by the fourth date (Figure 3). On the first date, the youngest internodes of the three wheats had a higher percentage of nitrogen than did the older internodes and this trend held throughout the growing period.

It was not possible to separate the pith and wall tissues from the internodes of the solid-stemmed varieties collected on June 20. However, the nitrogen content of the pith collected on the second date was similar to that of the wall from the corresponding internode. Thus it seems probable that the nitrogen content of the pith would be similar to that of the wall of the same internode at the earlier date.

Except for the samples collected on June 20, the nitrogen content of the three bottom internodes was generally similar for all varieties. Further, in the solid-stemmed varieties the nitrogen content of the pith was generally similar to that of the wall within any one internode. The largest difference occurred within the top internode, where the pith invariably contained a lower percentage of nitrogen than did the wall. However, the wall of the top internode of all varieties had a higher nitrogen content than did that of any of the lower internodes. During growth the percentage of nitrogen in the wall of the top internode was consistently higher in Golden Ball than that in the other two wheats. Apparently this was a reflection of the relatively slow development of Golden Ball. Furthermore, percentages of nitrogen for the three varieties at maturity differed only slightly. The percentages of nitrogen in the wall of the top internode were similar in Thatcher and Rescue and were intermediate between those of the wall of the top internode in Golden Ball and of the pith in the solid-stemmed varieties.

When the nitrogen content of the stem tissues was recalculated on a fresh weight basis certain relationships became apparent. For example, the percentage of nitrogen in Rescue pith was higher than that in Golden Ball pith. Furthermore, the nitrogen content of the tissues of the top internode of the wheats reached a peak during the growing season and declined with maturation. The peak for Golden Ball occurred at a later date than for either Thatcher or Rescue, presumably a reflection of the slow growing habit of Golden Ball. Peaks of comparable size were not evident for oats. The peaks appeared to have been the result of a proportionately small decline in nitrogen as a percentage of dry matter together with the general decrease in moisture content.

The percentage of nitrogen on a dry weight basis in sections of the top two internodes of Thatcher grown on irrigated land decreased during the growing period (Figure 4). On the dates immediately following the appearance of internode 4 and 5, respectively, more nitrogen was found in the bottom of the internodes than in the middle or top segments. As these internodes developed, the percentage of nitrogen decreased more rapidly

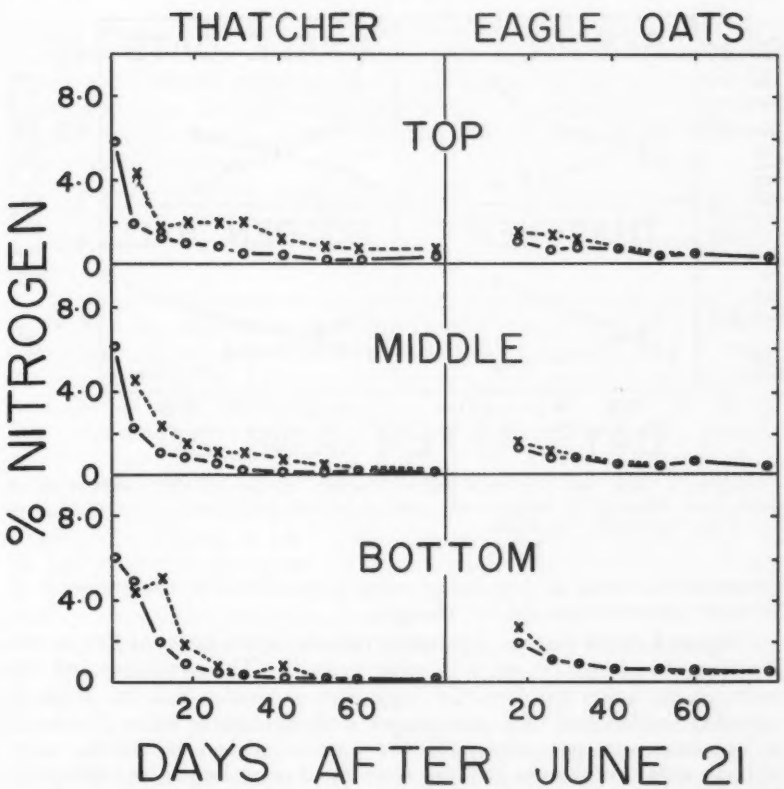

FIGURE 4. Percentage of nitrogen on dry weight basis in sections of internodes 4 and 5 from Thatcher wheat and Eagle oats grown on irrigated land. Because internodes 4 and 5 of Thatcher were undifferentiated on the first dates plotted the same value was used for each of the three sections within an internode. 0—0 internode 4; X---X internode 5.

in the bottom and middle than in the top, which resulted in a complete reversal of the nitrogen pattern during the following two weeks. The clearest illustration of this occurred in internode 5 of Thatcher; a similar but less evident change occurred in internode 4. The higher nitrogen level in the upper parts of the internodes, as in the upper internodes of the stem, may be the result of an upward migration of nitrogen to the developing heads.

Internodes of Eagle were fully formed when the first sample was taken and changes in nitrogen content of the different segments associated with early development were missed (Figure 4). In contrast to the nitrogen pattern in Thatcher, the percentage of nitrogen in Eagle throughout growth was highest in the bottom segments and lowest in the top segments of the two internodes. The percentage of nitrogen in the different parts of these

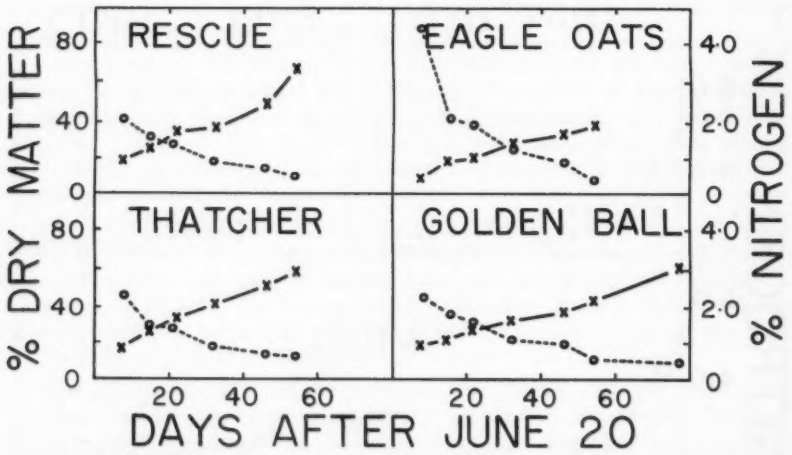

FIGURE 5. Dry matter content and percentage nitrogen on dry weight basis of nodes from three wheat varieties and one oat variety grown on non-irrigated land. X—X dry matter; 0---0 nitrogen.

internodes decreased during the growing period, but the relative order of nitrogen concentration did not change.

Figure 5 shows that the dry matter content of the nodes of Rescue and Thatcher was generally similar during growth. The percentage of dry matter in the nodes was lower in Eagle than in Golden Ball and lower in both than in Thatcher. The percentages of nitrogen in the nodes (Figure 5) of the wheats were generally similar but were lower than those in the nodes of Eagle oats. When the varieties were rated according to percentage of dry matter in the nodes, the order was the same as for percentage of dry matter in the internodes. The nodes of the wheats generally contained less dry matter and more nitrogen than the internodes. This difference resulted presumably from the fact that the nodes, in contrast to the internodes, contain the vascular tissues of both the leaf sheath and the internode (10). Comparison of the nodes and internodes of Eagle showed that the nodes were generally drier and contained more nitrogen than all but the top internode.

Relation of Tissue Composition to Sawfly Resistance

These results extend earlier findings (8) and permit the same conclusion: the percentages of moisture and nitrogen in the pith of the solid-stemmed resistant wheats were so similar to those of the stems of the hollow-stemmed susceptible variety that a quantitative nutritional deficiency of either moisture or nitrogen does not appear to be associated with resistance. It should be recognized, however, that the percentage of nitrogen in the walls of the solid-stemmed wheats or in the stems of Thatcher may be lower than that in the tissues which the larvae normally consume. Most of the feeding is in the region of the vascular bundles (3), which probably

contains larger amounts of readily available nitrogen than does the more sclerotized hypoderm. In contrast, the percentages of nitrogen reported for the pith probably represent the composition of the pith tissues that the larvae consume in the solid-stemmed varieties. Thus greater differences in nitrogen content than those recorded may exist between the pith and wall tissues actually consumed.

It is not yet known whether qualitative differences in nitrogen composition of wheat stems contribute to resistance. It has been concluded, however, that qualitative differences in carbohydrates are not contributing factors (9).

It has been observed that late in the growing period in most years more larvae die in the solid-stemmed resistant varieties than in the hollow-stemmed susceptible varieties (7). Based on this information and on our earlier chemical analyses (8) Holmes and Peterson have postulated that sawfly larvae die in the solid-stemmed varieties as a result of desiccation (4, 7). The larvae are known to migrate to the base of the stem late in the growing period (2) and, because there is a tendency for the two uppermost internodes to lose moisture most rapidly, it is probable that they move downward in search of moisture as ripening occurs. It would be expected that the downward movement is more difficult in solid than in hollow stems. If the pith delays the downward movement, the larvae could be in a dry environment much of the time which is unfavourable to them. The present results are consistent with this hypothesis.

It has been shown that a lower percentage of eggs hatched in the pith of Golden Ball than in the pith of Rescue, and also that eggs exposed to excess moisture for prolonged periods fail to hatch (6). Holmes and Peterson postulated that the excessive egg mortality in Golden Ball might result from a high moisture content in the pith at the time the eggs are laid. Present results support this hypothesis because the pith of Golden Ball does in fact contain more moisture (less dry matter) than does the pith of Rescue.

The quantitative data that were obtained for the oat plant do not explain the immunity of oats to the wheat stem sawfly. Larvae in oats generally die shortly after hatching (7), a time when these plants and susceptible wheat plants are not greatly different in either dry matter or nitrogen content.

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CROP ROTATIONS AND COMMON ROOTROT IN WHEAT[1]

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ABSTRACT

In a 5-year study of common rootrot of wheat in rotations at Indian Head, Saskatchewan, disease ratings were recorded three times each crop season and isolations were made to determine percentage of infection by *Helminthosporium sativum* and *Fusarium culmorum*. Wheat preceded by 0, 1, 2, 3, and 5 years of crops not susceptible to *H. sativum* had 68, 64, 37, 34, and 14 per cent, respectively, of *H. sativum* infected plants. Corresponding disease ratings for the same years were 28, 27, 22, 17, and 13 per cent, respectively. *F. culmorum* was isolated frequently from wheat in some of the rotations and it no doubt contributed to increased disease ratings. Oats in the rotation may be a factor in ensuring survival of this pathogen. Common rootrot in wheat may be lessened by using a comparatively long rotation.

INTRODUCTION

Common rootrot in Saskatchewan is responsible for losses ranging from 5 to 12 per cent (9, 10, 11). The dominant pathogen is *Cochliobolus sativus* (Ito and Kurib.) Drechs. ex Dastur whose conidial stage is *Bipolaris sorokiniana* (Sacc. in Sorok.) Shoemaker (*Helminthosporium sativum* P.K. and B.). The latter name, still in common usage by pathologists, has been retained throughout this paper. *Fusarium* spp., particularly *F. culmorum*, are also widespread and may be serious at times. Over a 5-year period Tyner (16) isolated *H. sativum* from 28 per cent of the wheat crowns and roots sampled in central Alberta and from 13 per cent in the Peace River area, whereas *F. culmorum* was isolated from 11 per cent in central Alberta and 31 per cent in the Peace River area. In Saskatchewan, a survey in 1958[3] revealed 51 per cent of the wheat plants infected with *H. sativum* in the southwest and 38 per cent in the northeast. *F. culmorum* was isolated from only 1.8 and 1.0 per cent of the plants in the two areas, respectively. However, isolations were from sub-crown internode material exclusively.

The literature contains numerous references citing control of various rootrots by means of rotation. Oats preceding the wheat has been reported helpful in controlling seedling blights and rootrot (1, 5, 8, 12, 15). Several workers, including Broadfoot (1), Henry (4), and Sanford (12), have reported some reduction in common rootrot as a result of summerfallowing. However, more recent work does not indicate lessened rootrot in the wheat crop after summerfallow (10, 11). The earlier investigations reporting control date back to the period when summerfallow implied the use of the mouldboard plough, and the control obtained is thought to be a result of burial of crop residues, surface soil, and spores (7). In current farm practice the mouldboard plough is seldom used; crop residues are maintained on the soil surface and no significant differences in incidence and severity of common rootrot are found as a result of the summerfallow year.

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[3]Ledingham, R. J. Annual Progress Rept., Research Sta., Can. Dept. Agr., Saskatoon. 1958. *Unpublished data.*

Chinn and Ledingham (2) found that under field conditions about 40 per cent of *H. sativum* spores were still viable after 2 years. It is thus apparent that a short rotation is likely to be only partially effective in eliminating *H. sativum* from soil.

The wheat crops of several rotations at the Experimental Farm, Indian Head, Saskatchewan, have been studied for the 5 years, 1956 to 1960, inclusive. In these rotations are wheat crops preceded by 0, 1, 2, 3, and 5 years of crops resistant to *H. sativum,* and summerfallow. Data obtained include incidence and severity of common rootrot at three dates each summer; fungi isolated from sub-crown internodes of wheat samples; populations of *H. sativum* spores in the plot soils, and finally incidence of disease in wheat seedlings grown in these soils in the greenhouse.

METHODS

This study is concerned mainly with three rotations, "C", "R", and "P", at the Experimental Farm, Indian Head, Sask. The crop sequence in these rotations is as follows: *rotation "C"*, summerfallow – wheat – wheat; *rotation "R"*, summerfallow – wheat – oats – hay – hay – hay – corn – wheat – oats; and *rotation "P"*, summerfallow – wheat (seeded down to sweetclover) – sweet clover (ploughed in late July after harvesting a crop of forage) – oats. The rotations are without replication and each plot is approximately 5 acres in extent (10 x 80 rods). The soil is classed as Indian Head clay to heavy clay and is reasonably uniform over the area involved. Rotations "C" and "R" have been in effect since 1912, and rotation "P" since 1934. The wheat crops were sampled in late June, the latter part of July, and at maturity each year. Four 100-plant samples were taken each time from the same area of each plot. Rootrot data, based on the incidence and severity of lesions on the sub-crown internode of the washed plants, were recorded in the laboratory. Disease ratings were obtained by use of the factors 2, 5, and 10 for slight, moderate, and severe ratings, respectively. The number of plants in each class was multiplied by its factor; the products were summed and converted to a basis of 100 plants.

Half-inch pieces of sub-crown internodes proximal to the crown, taken from 50 plants selected at random from each sample, were plated on acidified potato-dextrose agar. The pieces were surface sterilized by a 5-minute soak in 1:1000 $HgCl_2$, followed by precipitation of the residual mercury with a dip in a 2 per cent solution of sodium sulphide. The sub-crown internodes were washed thoroughly in Gooch crucibles with sterile water after each of the treatments. The plates were incubated at room temperature, and colonies of *H. sativum, F. culmorum,* and other readily recognizable fungi were recorded after about 10 days. In 1956 and 1957, *Fusaria* were recorded without differentiating species. In 1958, *F. culmorum* was recorded separately. However, in some cases it appears to have been confused with *F. avenaceum* and *F. acuminatum.* In 1959 and 1960, *F. culmorum* was recorded separately from other species. Soil samples were collected in September, 1959, and in May and September, 1960, from all of the plots in the rotations under study. Two collections were obtained

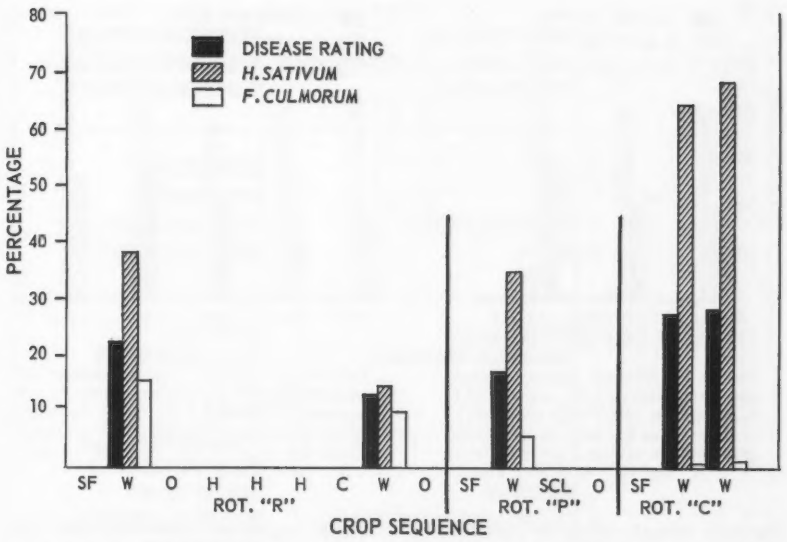

FIGURE 1. Average disease ratings and percentages of wheat plants infected with *H. sativum* and *F. culmorum* (1958-1959-1960 only) in rotations "R", "P" and "C" at Indian Head, *sampled three times each year, 1956-1960.* SF = summerfallow; W = wheat; O = oats; H = hay; C = corn; SCL = sweet clover.

from the surface 1.5 inches of soil of each plot. Each weighed about 30 pounds and was composed of about 150 small samples taken at random. After being brought into the greenhouse the soil collections were screened several times to ensure uniformity. The number and viability of *H. sativum* spores was determined by the flotation-viability method (3). In addition, soil from each plot was placed in quarter-gallon crocks and planted to wheat. Four replicates of each crop were allowed to grow for 30 days in the greenhouse at temperatures around 70°F. The plants were removed from the soil and disease ratings were obtained. Pieces of sub-crown internode were plated to determine the fungi involved.

EXPERIMENTAL RESULTS

Data presented graphically in Figure 1 show that disease ratings and percentages of wheat plants infected with *H. sativum* were influenced by the type of rotation. The same appears to apply to *F. culmorum.* Rotation "C" with 2 years of wheat followed by a year of summerfallow had a high disease rating and a very high incidence of *H. sativum* as determined by plating of sub-crown internodes. There was virtually no *F. culmorum* in this mono-crop rotation. Rotation "P", with wheat occupying a given plot only every fourth year and the interval being taken up with 1 year each of sweet clover, oats, and summerfallow, had a disease rating much lower than in rotation "C" and about half as many plants infected with *H. sativum.* *F. culmorum* was isolated from 6 per cent of the plants. Of

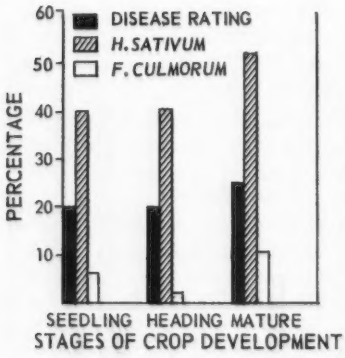

FIGURE 2. Average disease ratings and percentages of *H. sativum* and *F. culmorum* (1958-1959-1960 only) at three dates for each of the years 1956-1960, in wheat from rotations "P", "R" and "C" at Indian Head.

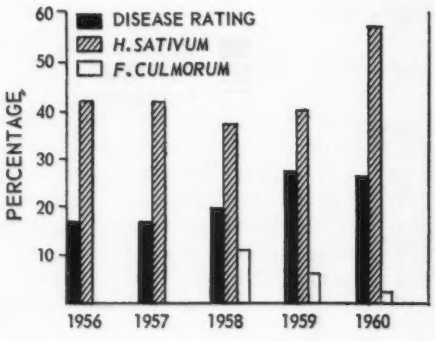

FIGURE 3. Average disease ratings and percentages of *H. sativum* and *F. culmorum* (1958-1959-1960 only) in wheat from rotations "R", "P", "C" at Indian Head for each of the years 1956-1960.

the two wheat crops in rotation "R", one is separated from the previous wheat by a year each of oats and summerfallow, while the other has a crop of oats, three of hay, and one of corn intervening. The disease rating and incidence of *H. sativum* was 13 and 14 per cent, respectively, in the latter, compared with 22 and 37 per cent in the former.

In Figure 2 are shown the disease ratings and incidence of *H. sativum* and *F. culmorum* in wheat plants at three different stages of development. It is of interest that the increases in disease rating and incidence of *H. sativum* from seedling stage to maturity were only slight. The incidence of *F. culmorum* was found to fluctuate quite widely from one year to the next and between stages of crop development.

In general, the disease ratings and percentages of *H. sativum* did not vary widely from year to year (Figure 3). In 1958, however, the disease ratings in the stubble wheat of rotation "C" were only 3, 10, and 9 per cent, in the seedling, heading, and mature crop stages, respectively (Figure 4). These values are very low, particularly since this wheat crop under normal circumstances would have maximum ratings. It will be noted that the seedling stage disease ratings were low in all of the wheat crops in 1958. However, with the exception of stubble wheat in rotation "C" the disease progressed rapidly and was at an above average level by crop maturity.

The number of *H. sativum* spores per gram of soil and the percentages of infected wheat plants grown in soil samples collected in September 1959 from each plot in the three rotations "R", "P", and "C" are shown in Figure 5. The presence of a wheat crop on any of the plots caused a substantial spore increase by mid-September. Spore numbers decreased during the winter by 22 per cent on the average, and the percentage of viable spores decreased from 86 in the fall to 58 in the spring. Spore counts made in the fall of 1960 showed a pattern similar to that of 1959, except that

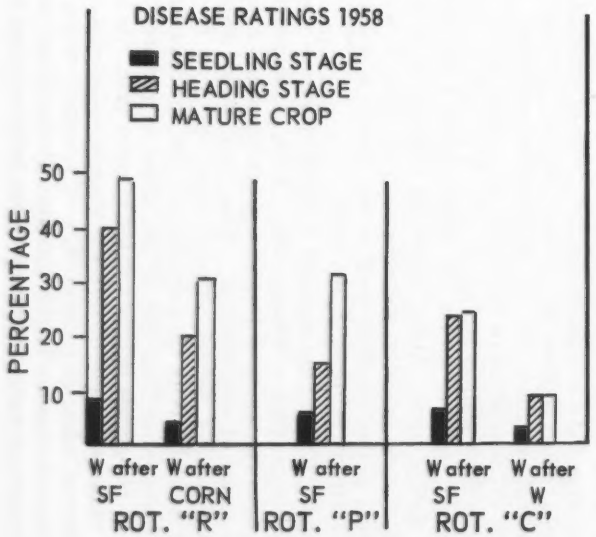

FIGURE 4

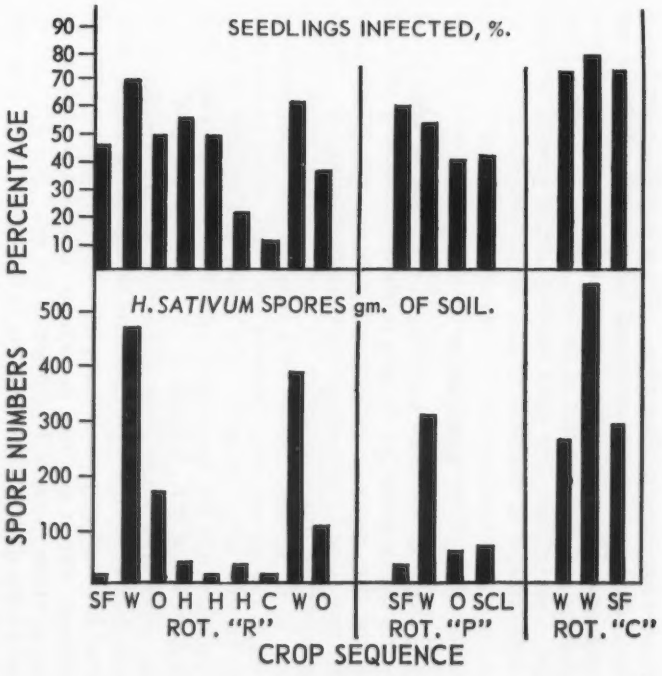

FIGURE 5

FIGURE 4. Disease ratings of wheat in rotations "R", "P", and "C" at three stages of development in 1958.

FIGURE 5. Numbers of *H. sativum* spores in soil collected in September, 1959, in rotations "R", "P", and "C" at Indian Head, and percentages of infected wheat seedlings grown in the soil samples in the greenhouse. SF = summerfallow; W = wheat; O = oats; H = hay; C = corn; SCL = sweet clover.

fewer spores were found in the wheat plots. This is very likely a reflection of the extreme drought of the summer and fall of 1960 which did not favour sporulation. The relationship between infected seedlings (determined by plating) and spore content of the soil (Figure 5) is not as close as anticipated. Nevertheless, low spore numbers are, in general, reflected in decreased percentages of infection.

DISCUSSION

The main object in this study has been to determine to what extent common rootrot can be controlled by crop rotation. Laboratory and field studies indicated that spores of *Helminthosporium sativum* persisted for a considerable time (2 to 3 years) in soil before losing viability (2). Spores of *Fusarium culmorum* probably survive for a much shorter period in soil than do those of *H. sativum*. Stover (14) reported that when spores of *F. oxysporum* f. *cubense* or other *Fusarium* spp. were added to soil they were seldom detected after 6 months. The author in a preliminary study found that comparatively few spores of *F. culmorum* remained viable after 3 months in moist soil at 0°F., whereas at 70°F. survival was good.

The two consecutive wheat crops of rotation "C" are preceded by 1 year of summerfallow. In this report the term summerfallow means "trash summerfallow" in which crop residues are retained in the surface layers of soil. It is obvious that at all times in rotation "C" there is likely to be an abundance of viable spores of *H. sativum* in the soil. In 3 years out of the 5 covered by this study the disease rating in the second wheat crop was higher than in the wheat after summerfallow. However, the ratings were high in both crops and it is apparent that the 1 year of summerfallow cannot be classed as a control measure for common rootrot. Indeed, the extensive surveys reported by Sallans (10, 11) support this contention. A survey in 1952 showed 192 summerfallow fields with an average disease rating of 7.2, and 57 stubble fields with an average of 6.3. A comparable survey in 1958 gave an average rating of 9.0 on 190 summerfallow fields and a rating of 10.1 on 52 stubble fields. Many factors are interdependent in rootrot infection and of these inoculum abundance is only one. It seems probable that, beyond a modest inoculum level in soil, rootrot is not increased substantially by further increases of inoculum.

In rotation "R" the crop sequence is summerfallow – wheat – oats – hay – hay – hay – corn – wheat – oats. Thus, the summerfallow wheat of this rotation was separated from the preceding wheat crop by a crop of oats and a year of summerfallow. This 2-year period should have had some effect in rootrot control, although many spores of *H. sativum* will survive this interval. With the exception of 1958, the disease rating of this wheat crop was lower than that of the wheat in rotation "C", and the percentage of *H. sativum* isolates was lower every year. As already mentioned, the extreme drought of 1958 was thought to be responsible for the light infection of wheat in rotation "C". The stubble wheat particularly had very low soil moisture in this rotation. It should not be overlooked that *F. culmorum* appeared commonly in the isolations from rotation "R".

It occurred, in fact, about half as frequently as *H. sativum.* No doubt it was responsible for some of the rootrot.

A comparison of the percentages of wheat plants yielding isolates of *H. sativum* in the different rotations is interesting. In the summerfallow wheat of rotation "C", 64 per cent of the plants yielded *H. sativum,* whereas in the summerfallow wheat crop of rotation "R" 37 per cent yielded *H. sativum.* The wheat of rotation "C" was separated from the previous wheat crop by a year of summerfallow, whereas in rotation "R" there was a crop of oats in addition to the summerfallow. The second year wheat in rotation "C" had 68 per cent of the plants infected with *H. sativum,* whereas rotation "P" with 3 years, and the wheat after corn in "R" with 5 years between wheat crops, had percentages of 34 and 14, respectively. Thus, the wheat crops of these rotations, preceded by 0, 1, 2, 3, and 5 years of crops not subject to attack by *H. sativum,* yielded wheat plants with 68, 64, 37, 34, and 14 per cent, respectively, of *H. sativum* infections.

The relative abundance of *F. culmorum* in rotations "R" and "P" is possibly related to the presence of oats in these rotations. This was partially confirmed by collecting soil from the rotations in September and seeding them to wheat in the greenhouse. Without exception, the wheat grown in soil that had produced a crop of oats in 1959 had a considerable number of infections due to *F. culmorum,* whereas the soils that were 2 or more years away from oats had little or no *F. culmorum.* It seems likely that *F. culmorum* does not survive well in the Indian Head soils in the absence of a suitable host. The spores probably die out almost completely over winter and the organism is carried on by the fungus present in invaded host tissue.

Counts of *H. sativum* spores in soils of the Indian Head rotations reveal very well the capability of this pathogen to produce an abundance of spores each time wheat is grown. The decline in spore numbers is relatively slow, and, in addition, spore populations of 20 to 50 per gram of soil cause considerable infection. Thus, a decline in spore numbers, such as occurs in the first and second years after wheat, is not likely to be reflected in lowered disease. A sharp decline in disease appears to come only after spore numbers drop to a low level. Even oats, regarded as highly resistant to *H. sativum* (13), has shown evidence, not only in this study but as well in a rotation presently under investigation at Swift Current, Saskatchewan, and in a greenhouse experiment, of supporting the fungus sufficiently to be undesirable as an alternate crop in the rotation. In this connection Kozyreva (6) has reported wild oats (*Avena fatua*) as a facultative host and an important source of infection of *H. sativum.* On the other hand, in the hay plots, a mixture of brome [a known host of *H. sativum* (13)] and alfalfa, spore counts dropped to a low level and there was little evidence of new spores being produced.

Sufficient information is not available to indicate whether a few infections on a plant may be as serious as many infections. Numerous infections of the seedling, together with other adverse conditions, may cause thinning of the crop. However, it has been observed in this study that

frequently disease ratings at crop maturity were as high in plots with moderately low spore numbers as in those with an abundance of spores.

A considerable measure of common rootrot control may be achieved through crop rotation but a comparatively long rotation is required.

ACKNOWLEDGEMENTS

The author wishes to thank E. V. McCurdy, Experimental Farm, Indian Head, Sask., for permission to study the rotations, and Mrs. J. Key, Canada Department of Agriculture Research Station, Saskatoon, Sask., for aid in recording and plating.

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VARIABILITY WITHIN ISOLATES OF *VERTICILLIUM ALBO-ATRUM* FROM POTATO[1]

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ABSTRACT

A monospore analysis of fresh isolates of *Verticillium albo-atrum* Reinke & Berthold from eight locations and six varieties of potato in Prince Edward Island, showed that, with one exception, the mass isolates were composed of morphologic forms distinguishable in growth habit and in capacity to form dark resting mycelium. One chief component, resembling the parent form, was present in all isolates. Pathogenicity trials in the greenhouse showed that the monospore forms differed in virulence and that virulence was not associated with the capacity to form dark, resting mycelium. Some monospore forms were avirulent, and one series was found that was equally pathogenic on both the highly susceptible variety Irish Cobbler and the field resistant variety Houma.

INTRODUCTION

Verticillium albo-atrum Reinke & Berthold, distinguishable by the formation of dark, resting mycelium and the absence of pseudosclerotia, has been found to be the most pathogenic and predominant form of *Verticillium* in some major potato-growing areas of Eastern Canada (4). Among potato varieties tested for resistance to this species the variety Houma has been reported to be highly resistant, although occasionally plants of this variety succumb rapidly to the disease (1). Such sporadic susceptibility in an otherwise highly resistant variety suggests that strains of *Verticillium* may exist, or will arise, to attack resistant varieties of potato.

Cultural and pathogenic variability has been reported to occur within mass isolates of *Verticillium albo-atrum* from potato (4, 5). The purpose of the present work was to study the variation in form and in pathogenicity that may exist within a group of fresh isolates of this fungus.

MATERIALS AND METHODS

Stem tissue of wilted plants obtained from naturally infected fields was surface sterilized and plated. Pieces from which the fungus grew in pure state were transferred to agar slants and from the mass isolates so obtained a monospore analysis was carried out. Spore suspensions were made to contain, by haemocytometer count, about 100 spores/millilitre and aliquots of ½ millilitre were pipetted evenly on to the surface of freshly poured, 3 per cent potato dextrose agar. Examination of plates, and of spore suspensions prior to plating, showed no evidence of any clumping of spores. In 2 or 3 days at 70°F. colonies became visible on each plate and after 6 days these were counted, classified for type, and a number, usually 100, transferred to bottle slants for further observation. All comparisons were made using potato dextrose agar.

Plants used in pathogenicity trials were grown in sand watered with Hoagland's nutrient solution until a suitable size for transplanting, then uprooted at time of inoculation and transplanted to 4-inch pots containing

[1]Contribution No. 102, Experimental Farm, Research Branch, Canada Department of Agriculture, Charlottetown, P.E.I.

a 3:1 mixture of compost and sand. Potato plants were grown from single eyes and the seed pieces detached before inoculation.

Inoculations were carried out by dipping roots in inoculum immediately prior to transplanting. All inoculum consisted of spore suspensions made from 2- to 3-week-old cultures. Before use, the suspensions were filtered and adjusted to a uniform density of 2 to 3 x 10^6 spores/millilitre. For field planting, potato seed pieces were dipped in spore suspensions of the desired cultures and planted on land previously in sod and where Verticillium wilt had never been observed.

EXPERIMENTAL RESULTS

Eight mass isolates of *Verticillium* were obtained from different potato stocks in widely separated locations. After 2 to 4 weeks growth in culture all the isolates developed dark, resting mycelium characteristics of *V. albo-atrum.* A monospore analysis, carried out with each isolate, showed that morphologically different forms existed within the mass isolates. These forms were usually distinguishable in the petri plates after 4 to 5 days. At that time some colonies were appressed with slight aerial mycelium and this form invariably turned dark after prolonged growth. It was designated 'WD'. Other colonies developed a fairly dense white aerial mycelium and did not darken after prolonged culture. This form was designated 'F'. After several months in culture the WD forms were typically dark and had a surface granulation which, microscopically, could be seen due to great numbers of mucoid droplets suspended on the conidiophores. The white, F, forms rarely showed any such granular appearance and usually retained more aerial mycelium. In addition to these two main forms, a few colonies appeared that were very slow growing 'D' or very mucoid 'M' in growth habit. Also, several colonies were encountered in the F series that produced a few scattered knots of darkened mycelium after periods of a year or more in culture. The proportion of the component forms occurring in the isolates studied is given in Table 1. The WD and F forms together constituted the bulk of the population in each monospore series. All the forms

TABLE I. — MORPHOLOGIC FORMS OCCURRING IN MONOSPORE ANALYSES OF FRESH ISOLATES OF VERTICILLIUM FROM POTATO

Isolate no.	Infected variety	Number colonies examined	Per cent of morphologic forms			
			WD	F	D	M
1	Irish Cobbler	1519	96.6	2.7	0.7	—
2	Saco	811	96.2	3.5	0.3	—
3	Sebago	1502	81.4	18.6	—	—
4	Keswick	5000	99.8	0.1	—	0.1
5	Sebago	1354	91.4	8.1	0.5	—
6	Sebago	214	92.0	7.5	0.5	—
7	Boone	1224	100.0	—	—	—
8	F5028[1]	2308	99.9	0.1	—	—

[1]Wilt-susceptible selection from the Potato Breeding Program, Fredericton, N.B.

produced typical fructification but the predominant form, WD, and the M cultures were usually more moist, with conidia commonly held in droplets at the tips of the verticils, or, in the case of the M cultures, forming a slimy surface.

Subculturing of these forms over a period of 3 years has shown no change in their general growth habit but in many of the WD cultures the capacity to form dark, resting mycelium was gradually lost. However, the initial separation of mass isolates into black and white components, which remain relatively stable, is in marked contrast to the variable morphology that results when mass transfers are made from isolates. In mass isolate transfer the sub-cultures may resemble the parent culture but often give black or white sectors and variations in darkening and mycelial development.

A second-generation monospore series was made with the WD and F forms of three of the isolates listed in Table 1. In every case the F forms gave rise only to white, F cultures. One WD culture gave rise to a like form but the other two gave colonies that represented a range in capacity to darken, with some colonies remaining white. In general the capacity to form dark resting mycelium was less in the second generation monospore series than in the first.

A characteristic sign of *Verticillium* infection on Irish Cobbler potato is the blackening of roots and stolons that becomes evident in the autumn after the plants have died. The capacity of six F and WD isolate forms to produce this blackening was studied in a field trial in which 140 plants were inoculated with each isolate. It was found that all the isolates behaved similarly in that less tissue blackening always occurred with the F than with the WD forms. The average percentages of plants showing wilt and blackening were as follows:

Isolate form	Per cent wilt	Per cent blackening
F	21.2	1.9
WD	24.9	11.7

A culture representative of the WD and F series of six of the isolates listed in Table 1 was chosen for further study. These cultures were tested for pathogenicity on tomato (var. Earliana), eggplant (var. Black Beauty) and potato (var. Houma and Irish Cobbler). Ten plants of each variety were inoculated respectively with each culture. Host species were chosen because of their known susceptibility, except for the resistant potato variety Houma, to mass isolates of *V. albo-atrum*. Wilt severity for three isolates representative of the group is given in Table 2. Pathogenicity was apparently not related to the capacity to form dark resting mycelium. It is notable that one culture, 1F, was slightly pathogenic on the potato variety Houma and another, 1WD, was avirulent on three of the four hosts.

TABLE 2.—REACTION OF EGGPLANT, TOMATO AND TWO VARIETIES OF POTATO TO MONOSPORE FORMS OF SEVERAL VERTICILLIUM ISOLATES

Isolate no. and form	Wilt severity[1]			
	Eggplant[2]	Tomato[2]	Potato-Houma[3]	Potato-Irish Cobbler[3]
F form				
1	**	**	*	**
2	***	**	0	***
3	**	*	0	**
WD form				
1	0	0	0	*
2	***	**	0	***
3	*	**	0	**

[1] 0 none; * slight; ** moderate; *** severe
[2] Disease readings after 16 days
[3] Disease readings after 33 days

In other, similar, trials using sub-cultures of the original monospore lines and also second-generation monospore series derived from them, it was found that isolate 1F remained pathogenic on the variety Houma. Other monospore lines were encountered that were only weakly pathogenic on Irish Cobbler potato. The slow growing form, D, and the mucoid form, M (Table 1) were found to be completely avirulent when tested on eggplant and potato.

To further confirm the virulence of the 1F isolate on the variety Houma, two trials were carried out. In the first trial, 25 cultures were chosen at random from a large monospore population of the original 1F culture. All were white and similar to the parent form. Each culture was used to inoculate 20 Houma plants. Wilt began to appear in 8 weeks and progressed rapidly, with all the cultures proving virulent. In the second trial, several re-isolates from plants of the first test, together with the original 1F isolate, were used. Thirty Irish Cobbler and Houma plants were inoculated with each culture. Wilt began to appear in 6 weeks and progressed steadily. Once again, all isolates proved to be virulent. The pathogenicity of these isolates, (Table 3) was undoubtedly as great on the wilt-resistant variety Houma as on the wilt-susceptible variety Irish Cobbler.

In all the pathogenicity trials, a re-isolation of the fungus was made from the hosts used. It was found that the organism was recoverable from both resistant and susceptible plants but to a lesser degree, except for isolate 1F, from the resistant potato variety Houma. The WD and F monospore forms generally retained their distinctiveness (Figure 1), although not to an equal degree. The F forms were always recovered unchanged but WD forms sometimes apparently lost the capacity to form dark mycelium or retained it to a limited degree, giving colonies that ranged from jet black to white.

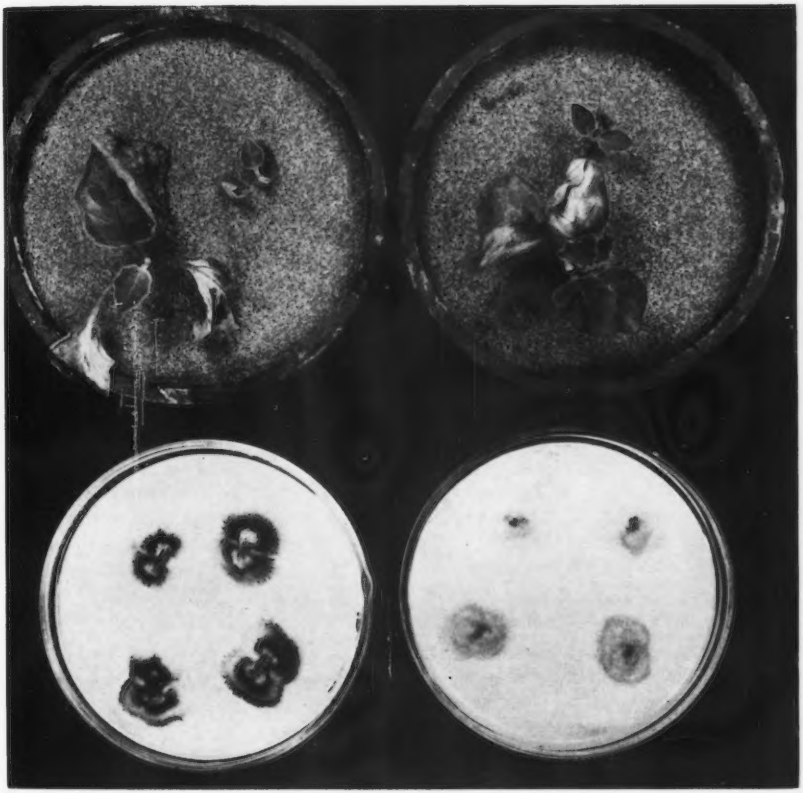

FIGURE 1. Wilt in eggplant caused by two monospore forms of *Verticillium albo-atrum*. Upper: *left* – WD form; *right* – F. form. Lower: Recovery of respective forms from infected tissue.

TABLE 3. — THE INCIDENCE OF WILT IN THE POTATO VARIETIES HOUMA AND IRISH COBBLER WHEN INOCULATED WITH CERTAIN MONOSPORE LINES OF VERTICILLIUM

Culture		Wilt severity[1]		
		Trial 1	Trial 2 re-isolates from Trial 1	
		Houma	Irish Cobbler	Houma
Sub-monospore lines	IF	**	***	**
	A	***	**	**
	B	***	***	***
	C	***	***	***

[1] * slight; ** moderate; *** severe

DISCUSSION

In most studies of pathogenic *Verticillia* it has been noted that cultural variation abounds. The occurrence of several homotypes in monoconidial cultures of *Verticillium* was reported by Hansen (2) in his study of the dual phenomenon in the imperfect fungi, and more recent work has shown that distinct component forms may be derived from mass isolates of both pseudosclerotial and non-pseudosclerotial *Verticillia* (3, 4). In the present study this same general pattern of variability was found. Since the conidia of this fungus are almost invariably uninucleate (4, 5), it would be expected that single spores would give rise to homocaryons that would remain stable except for subsequent mutation or changes brought about by environment. The behaviour of the mass isolates studied here indicates that they are a mixture of several homocaryons with two morphologic types predominating. In some cases isolates also apparently contain factors for pathogenicity which are possible to segregate by isolating monospores.

The capacity to form dark, resting mycelium is a feature separable by monospore culturing and thus explicable, at least to a degree, on the basis of heterocaryosis. However, the stability of this morphologic feature is greatly influenced by environment. As found in other work (4), fresh isolates from naturally infected tissue always formed dark, resting mycelium and were identifiable as *V. albo-atrum* when cultured under ordinary laboratory conditions. All mass isolates and some of the WD monospore lines gradually showed less capacity to form dark mycelium when sub-cultured over long periods. On the other hand, some F forms, which remained white in culture, developed some dark mycelium on infected plants in the field. These phenomena suggest that environmental conditions may suppress or modify the genetic factors that initiate the formation of dark mycelium.

Although variation in the pathogenicity of monospore lines of *V. albo-atrum* from potato has been reported (5), no evidence was found of specific pathogenicity on an otherwise resistant variety. It seems evident from the present work that pathogenic forms, capable of attacking resistant varieties may exist within mass isolates of *Verticillium* and so account for the occasional infection of resistant plants in the field.

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INHERENT DIFFERENCES IN DEPTH OF CROWN IN WHEAT AND BARLEY[1]

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ABSTRACT

Varieties of spring wheat and barley were found to differ considerably in the length of subcrown internode and consequently in the depth of crown formation in the soil. Single plant selections within varieties showed no significant variations in depth of crown. The depth of crown appeared to be a heritable character in randomly selected lines from the cross Thatcher x Reward, the former parent tending to form deep crowns and the latter shallow crowns. Possible physiological and pathological effects of depth of crown are discussed.

INTRODUCTION

Two observations drew attention to varietal differences in the depth of crown formation in spring wheats. In recording lesions caused by the root disease fungi, *Helminthosporium sativum* P.K. and B. (*Bipolaris sorokiniana* [Sacc. in Sorok. (Shoemaker)], and *Fusarium* spp. on a group of wheat varieties, Thatcher was observed to have shorter subcrown internodes than the other wheats. This characteristic appeared to result from the formation of Thatcher crowns at a deeper level in the soil, rather than from a shallower placement of the seed. The second observation was made in connection with a late spring frost. Varieties such as Rescue and Regent were killed to the extent of 80 to 90 per cent of the plants, whereas Thatcher in the same and nearby fields had about 10 per cent of the plants killed. The leaves of all varieties had been killed by the frost. Examination showed that the crowns of surviving plants of all varieties were more deeply placed than those of the dead plants. The high rate of survival of Thatcher was apparently related to its having more deeply placed crowns than the other varieties.

Webb and Stephens (5) found depth of crown formation in winter wheats to be influenced by variety, temperature, and depth of seeding. In general, winter wheats formed crowns more deeply than did spring wheats, and hardy winter wheats formed crowns deeper in the soil than did non-hardy varieties. Taylor and McCall (4) found that the length of the subcrown internode in wheat was increased by deeper seeding and by high temperatures.

A preliminary comparison was made of 22 varieties of spring wheat in the greenhouse in midwinter. The varieties, sown 76 millimetres deep in soil, showed a range in depth of crown formation from 15 millimetres in Reward to 31 millimetres in Canus and Thatcher. This paper reports on further varietal comparisons made in the field and greenhouse as a preliminary step in studies of depth of seeding and depth of crown in relation to common rootrot caused by *H. sativum* and *Fusarium* spp.

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MATERIALS AND METHODS

In the field experiments, plants of different varieties of spring wheat were grown in grids constructed of wooden boards 4 inches wide and ⅜ inch thick. The cells of the grids were usually 8 x 8 inches and 4 inches deep. The number of cells per grid in different experiments were 25, 36 or 49. The grids were placed in the field by removing about 3 inches of top soil, and setting the grids on the freshly exposed surface. The seed, 20 to 30 of one variety to a cell, was placed on the soil and covered with equal volumes of soil per cell to a uniform depth of 76 millimetres. The soil was lightly tamped. Greenhouse tests were made in beds of soil in a similar manner, the cells of the grids consisting of horticultural cubes with open bottoms.

The experimental design included two replicates of a simple partially balanced lattice arranged in two squares, 5 x 5, 6 x 6 or 7 x 7. The varieties were arranged in the squares in blocks as described by Cochran and Cox (1).

Data were taken in greenhouse tests 4 to 5 weeks after seeding and in field tests usually after the plants were in head. Measurements were taken on the depth of the crown and length of the subcrown internodes of 10 randomly selected plants from each cell of each grid. The data were analysed and Duncan's multiple range test of significance (2) was used.

TABLE I.—DEPTH OF CROWN BELOW SOIL LEVEL OF 49 WHEAT VARIETIES

Variety	Depth of crown (mm.)[1]	Variety	Depth of crown (mm.)[1]
Brownie	28	Blue Chaff	42
Reward	31	Redman	42
Baart	33	*T.V. rigidum*	43
Regent	33	Supreza	43
H-44-24	34	Kota	43
Florence[2]	34	Garnet	44
Chinese Spring	34	Marquis	44
Sevier	35	Lee	45
Prelude	35	Sea Island	45
Apex	35	Red Fife	46
Crown	35	Stanley	47
Ruby	35	Kanred	47
White Russian	36	Cadet	47
Pioneer	36	Kenya 9	47
Propo	36	Kenya 12	48
Selkirk	36	Lake	48
Hope	38	Rival	51
Merit	39	C.T. 510	53
Blue Ribbon	39	Premier	54
McMurachy	40	Huron	55
Saunders	40	Thatcher	55
Red Bobs	40	Canus	59
Preston	40	Reliance	59
Ladoga	41	Double Cross	65
Rescue	41	—	—

[1]S.E. of a variety 2.76 millimetres
[2]Doubtful identification

RESULTS

Differences between Varieties

Table 1 shows variations in depth of crown between 49 varieties of spring wheat grown in two replicates of a 7 x 7 lattice in the field at Saskatoon. Since the average depth of seeding was 76 millimetres the difference between this value and the depths of crown represented the length of the subcrown internodes of the varieties. The maximum depth of crown for Double Cross, a sister line to Thatcher, was 65 millimetres, 37 millimetres greater than that for Brownie.

A second experiment included a number of barley varieties, and some wheats not used in the first test. Four wheat varieties, Reward, McMurachy, Thatcher and Canus, were common to both tests. The second test was planted May 20, 11 days later than the first one, when the soil was several degrees warmer. The depth of seeding averaged 71 millimetres. Table 2 shows the depths of crown in millimetres for wheat and barley varieties.

There was a tendency for crowns of wheat in this test to be shallower than in the first test. The higher soil temperatures of the second experiment appear to have favoured greater elongation of the subcrown internodes, and, therefore, the crowns tended to be closer to the soil surface. The four varieties of wheat common to both tests fell into the same order each time. As in the first experiment the wheat varieties showed a wide range of variation in depth of crown. Of the durums in the test Pelissier, Carleton and Mindum had deeply placed crowns, while Golden Ball and Iumillo were intermediate in relation to the common wheats. There was also a significant variation in depth of crowns among the barley varieties.

TABLE 2.—DEPTH OF CROWN BELOW SOIL LEVEL OF 16 WHEAT AND 19 BARLEY VARIETIES

Wheat variety	Depth of crown (mm.)[1]	Barley variety	Depth of crown (mm.)[1]
Reward	30	Newal	19
Yaroslav	30	Titan	24
Kenya 10	31	Sanalta	25
Red Russian	31	Law	26
McMurachy	32	Vantage	28
Bobs	33	Lion	28
Kenya 8	35	Rex	30
Golden Ball	36	Hannchen	31
Iumillo	36	O.A.C. 21	31
Kenya 11	36	Olli	32
White Fife	38	Double Cross	32
Pelissier	40	Regal	33
Thatcher	40	Warrior	33
Canus	41	Plush	35
Carleton	44	Velvon	37
Mindum	49	Trebi	37
—	—	Glacier	37
—	—	Prospect	38
—	—	Montcalm	45

[1]S.E. 2.9 millimetres

In 1954, a retrial was made of 25 varieties chosen from the 49 of the first experiment. The test was conducted in the same way; however, the depth of seeding was only 66 millimetres. The depth of seeding, nevertheless, was sufficient to obtain a wide range in depth of crown. A correlation coefficient of 0.893 was found between the 2 years' results on the 25 varieties.

Fifteen of the varieties of wheat common to the 1953 and 1954 wheat tests were also present in the preliminary greenhouse trial mentioned in the introduction. The coefficients of correlation between the preliminary test and the two field tests were respectively 0.835 and 0.885. There is strong evidence, therefore, to suggest that the varietal differences in depth of crown formation are largely genetically controlled.

Purity of Varieties with Respect to Depth of Crown Formation

Measurements were made on depth of crown of randomly selected single plant lines of Thatcher, Lake, and Sevier. Depth of crown in 36 lines of Thatcher ranged from 40 to 48 millimetres, with a standard error of 2.57 millimetres; in 25 lines of Lake from 36 to 44 millimetres, with a standard error of 1.66 millimetres, and in 36 lines of Sevier from 29 to 41 millimetres, with a standard error of 2.47 millimetres. With the possible exception of three lines of Sevier there was no evidence in these tests to indicate heterogeneity for depth of crown. A greenhouse test of the six lines from each variety that showed greatest deviation from the means confirmed this finding. There was no evidence that such deviations represented an inherent difference in depth of crown formation between the lines of a given variety.

Inheritance of the Depth of Crown Character

Since Thatcher and Reward form deep and shallow crowns respectively, a comparison of depth of crown was made of a number of randomly selected lines from a cross between them. The lines were in the F 9 and F 10 generations. The field trial was seeded in two 7 x 7 lattices, the seed being covered to a depth of 76 millimetres. Measurements of the depth of crowns were taken about mid-June, 4 weeks after seeding. The data are summarized in Table 3.

The depth of crown of the 49 lines from the cross ranged between those of the Thatcher and Reward parents. The multiple range test not only indicates that the upper and lower portions of the distribution of lines (Table 3) could not be distinguished from Reward and Thatcher, respectively, but also that an intermediate group of lines differed significantly from both parent varieties.

A greenhouse experiment in an 11 x 11 partially balanced lattice design (three replications) included 96 lines of Thatcher x Reward, 13 samples from a bulk lot of Reward, and 12 similar samples of Thatcher. The seed was covered to a uniform depth of 76 millimetres. During emergence the seedlings were given artificial light at night in addition to normal daylight.

The results of this test were very similar to those of the previous test. There were 39 lines from the cross common to both experiments. The coefficient of correlation based on these 39 lines was 0.735. There was.

TABLE 3.—DISTRIBUTION OF RANDOM LINES FROM THATCHER x REWARD CROSS AS TO DEPTH OF CROWN IN A FIELD TRIAL

Depth of crown (mm.)	Thatcher	Thatcher x Reward	Reward	Multiple range test[1]
26	—	2	—	
27	—	4	1	
28	—	1	—	
29	—	2	—	
30	—	4	—	
31	—	5	—	
32	—	3	—	
33	—	6	—	
34	—	2	—	
35	—	3	—	
36	—	4	—	
37	—	3	—	
38	—	2	—	
39	—	2	—	
40	—	2	—	
41	—	2	—	
42	—	0	—	
43	—	1	—	
44	—	1	—	
45	1	0	—	

[1]S.E. of a line is 2.54 millimetres

therefore, confirmation that some lines resembled the Reward parent, some the Thatcher parent, and other lines were intermediate between the parents in depth of crowns. These experiments indicate that lines from the cross have segregated for depth of crown and that selections of lines similar to each parent or intermediate between them is possible.

DISCUSSION

Depth of crown may be of importance not only in winter wheats (5) but also in spring wheats and barley varieties. It may be significant that many of the more useful wheat varieties on the dry plains of Western Canada, such as Thatcher, Canus, Lake and Mindum, form deep crowns, or that such formerly useful wheats as Red Fife, Marquis, Garnet, Redman, Rescue, Red Bobs, and Saunders are intermediate in depth of crown; while, on the other hand, less useful varieties, such as Reward, Brownie, Regent, Prelude, and Ruby, form shallow crowns. Among the most useful barley varieties Montcalm, Hannchen, O.A.C. 21, and Olli were found to have deep or intermediate crowns, while the shallow crowned varieties Newal, Titan, Sanalta and Vantage did not prove to be widely adapted to conditions in Western Canada. The reasons for such a relationship may be several, and are largely a matter for speculation till more is known.

If sufficient evidence is built up for the value of deeply placed crowns in spring wheat, for example in relation to the root diseases, it appears that plant breeders will be able to select for this character when deep-crowned varieties, such as Thatcher, are used in their crosses. Selection would best be done on lines seeded 2.5 to 3 inches deep under field conditions.

The observations on survival of wheat varieties from a late spring frost suggest that environmental factors are of importance. Though Thatcher may be inherently more resistant to frost damage than Rescue, the loss of the leaves in both varieties indicates that this may not have been the main factor determining survival. The loss of shallow-crowned individuals in both varieties, and survival of deep-crowned individuals suggests that the depth of crowns may have determined which plants would be completely killed and which would survive. Survival in this case would involve the regrowth of leaves from the crown, or from that portion of new leaves enclosed in the sheaths of the first and second leaves between soil level and the crown. The longer this section of new leaves, the more likely the plants are to survive.

Though severe injury to wheat from spring frost is not very frequent in Saskatchewan, injury caused by high temperatures and drought are not uncommon and infections of various tissues and organs below ground level by *Helminthosporium sativum* and *Fusarium culmorum* are very common. Severe drought during early to mid-June, with complete drying of the top 2 or 3 inches of soil, interferes with normal development of crown roots. Under such conditions, the shallower the crowns are in the soil the less is the likelihood that crown roots can be established. Not only do infections of wheat by *H. sativum* tend to be more severe under somewhat droughty conditions than when soil moisture is optimum (3), but the position of the crown, which determines the length of the subcrown internode, may influence the infections of both structures. Evidence on the latter point is being gathered for presentation in another paper. Further work is also being done on the influence of environmental factors in determining the crown level in wheat.

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VARIATION IN THE SENSITIVITY OF ISOLATES OF *VENTURIA INAEQUALIS* (CKE.) WINT. TO FUNGICIDES[1]

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ABSTRACT

Fifty isolates of *Venturia inaequalis*, from diseased apples of several varieties collected in sprayed orchards on various fungicide programs and in unsprayed orchards, were tested for their sensitivity to captan, glyodin, dichlone, phenyl mercury acetate, copper sulphate and sulphur. The isolates were sensitive to a wider dosage range of sulphur than of other fungicides but there was no indication that, in nature, *V. inaequalis* had developed strains resistant to the fungicides.

INTRODUCTION

The repeated use of some insecticides against insects and some antibiotics against bacteria has resulted in the development of resistant strains but there has been little evidence that fungi become resistant to fungicides in nature. Taylor (14) obtained results which indicated that strains of *Physalospora obtusa* (Schw.) Cooke resistant to Bordeaux mixture developed in orchards where this fungicide had been used for several years. It has been suggested (5) that increased difficulty in controlling *Phytophthora infestans* (Mont.) DeBary on potato with Bordeaux mixture was due to an acquired resistance by the fungus to copper. Wollman (15) found differences in sensitivity to fungicides between different isolates of pathogenic fungi. There are numerous examples of the development *in vitro* of resistance by plant pathogenic fungi to fungicides (3, 4, 5, 6, 7, 8, 10, 11, 12, 13).

In Nova Scotia, copper and sulphur fungicides have been used for many years to control apple scab caused by *Venturia inaequalis* (Cke.) Wint. More recently, they have largely been replaced by organic fungicides. Investigations were carried out to determine if, in nature, *V. inaequalis* has developed strains resistant to these different fungicides. The results obtained are given in this paper.

MATERIALS AND METHODS

During the growing season of 1957 isolations of *V. inaequalis* were made from diseased apples of several varieties from orchards on various fungicide programs and from unsprayed orchards. Most of these apple orchards were located in the Annapolis Valley, where fungicides have been used for many years, but some were in areas where it is unlikely that fungicides have ever been used. Monoconidial transfers were made from the original isolations and the cultures were maintained on P.D.A.

Toxicity to the isolates of *V. inaequalis* was determined by the test-tube dilution technique for use with the slide-germination method essentially as described by the American Phytopathological Society, Committee on Standardization of Fungicidal Tests (1, 2). Conidia for the assays were grown as follows: A small portion of an agar culture was ground up in

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sterile water and suspended in a 10 per cent malt extract solution. Ten millilitres of this mixture were added to a sterile 9-centimetre Petri dish with a layer of cheesecloth in the bottom and a side wall liner of cellucotton. The latter acted as a wick and served to maintain even moisture in the dish without any excess of free liquid. The plates were incubated for 15-25 days at 18°C. Because there was variation in the number of conidia produced by different isolates of *V. inaequalis* it was often necessary to carry out assays with a lower concentration of conidia than that recommended (1).

The fungicides used were captan, glyodin, dichlone, phenyl mercury acetate, copper sulphate and sulphur. Technical grade samples of the first four were obtained from manufacturers*. Stock solutions of captan, glyodin and dichlone were prepared in absolute ethyl alcohol; phenyl mercury acetate and copper sulphate in water; and sulphur in acetone. When added to water the latter gave a colloidal sulphur solution (9). The potency of these stock solutions was checked at regular intervals. In the assays the solvents were at concentrations that had no effect on the germination of conidia. Following preliminary assays to determine the dosage range for each fungicide, each isolate was assayed against each fungicide on 2 different days and the results were recorded as the average of the two assays. Fifty isolates of *V. inaequalis* were assayed against each fungicide. These isolates were from 17 varieties of apple collected in 22 orchards.

RESULTS AND DISCUSSION

Only a few assays of each fungicide gave data adaptable to dosage-response curves, so it was not feasible to compare the sensitivity of the isolates by their ED50 values. The data in Table 1 were selected to show the variation in the per cent inhibition of germination of the isolates by each fungicide. The data in Table 2 give the number of isolates showing complete inhibition of germination at each fungicide dilution.

The isolates varied somewhat in their sensitivity to the various fungicides, but there was no indication that any isolate had acquired resistance to any of the fungicides. Part of the variation may have been caused by the concentration of spores used in the assays. With isolate 19-57, for example, the spore concentration was only 7000 per millilitre and this isolate was more susceptible than the other isolates to sulphur, copper sulphate and captan (Table 1). Any indication of resistance does not appear to have been caused by the previous spray program. Isolate 19-57 was from an orchard in which sulphur fungicides had been used for many years. Isolate 4-57 was from an orchard that had never been sprayed with sulphur but, in the assays, was quite resistant to this fungicide. Isolate 14-57, from an orchard in which captan had never been used, showed some resistance to captan. The results in Table 1 indicate that 47-57 was somewhat resistant to dichlone. This isolate was from a glyodin-sprayed orchard. Isolates 34-57 and 85-57 appear to be resistant to phenyl mercury acetate. Both

*Captan, N-(trichloromethylthio)-4-cyclohexene-1,2-dicarboximide (California Spray-Chemical Corp., Eastern Research Lab., Moorestown, N.J.).
Glyodin, 2-heptadecyl-2-imidazoline acetate (Carbide and Carbon Chemicals Co., New York, N.Y.).
Dichlone, 2,3-dichloro-1,4-naphthoquinone (Naugatuck Chemicals, Elmira, Ont.).
Phenyl mercury acetate (Green Cross Insecticides, Montreal, Que.).

TABLE 1.—PER CENT INHIBITION OF GERMINATION OF DIFFERENT ISOLATES OF *V. inaequalis* AT VARIOUS DOSAGES OF FUNGICIDES

Fungicide	Isolate	Spores per ml. in assay in 1000's	Initial conc. p.p.m.	Dilution					
				0	½	¼	⅛	1/16	1/32
Glyodin	1–57	35	10	100	100	60	11	8	8
	4–57	50		100	95	87	21	9	2
	44–57	50		100	100	99	30	13	1
	93–57	50		100	100	100	40	17	3
Sulphur	1–57	35	10	100	100	100	96	64	21
	4–57	50		100	95	91	54	6	0
	19–57	7		100	100	100	100	100	64
	59–57	50		100	100	100	100	98	64
Copper sulphate	1–57	35	20	—	95	21	6	0	0
	19–57	7		—	100	98	21	2	0
	43–57	30		98	98	50	0	0	0
	59–57	50		100	100	54	15	3	1
Captan	1–57	35	5	100	100	87	20	12	2
	14–57	16		100	99	90	65	14	11
	19–57	7		100	100	100	100	15	3
	51–57	50		100	100	100	92	23	13
Dichlone	1–57	35	0.5	100	100	100	13	2	—
	11–57	50		100	98	75	35	0	0
	35–57	50		100	100	100	100	64	34
	47–57	50		100	98	76	7	4	4
Phenyl mercury acetate	1–57	35	0.05	100	100	91	55	9	8
	34–57	50		100	88	21	9	0	0
	50–57	50		100	100	100	81	33	22
	85–57	50		26	3	2	1	0	0

isolates were taken from a neglected orchard in which it is unlikely that mercury fungicides have ever been used. Isolate 1-57 was from a plot sprayed for several years with mercury fungicides. It showed no particular resistance to phenyl mercury acetate. Since there appears to be no correlation between previous spray treatments and the sensitivity of the isolates to the various fungicides, it is possible that naturally-occurring strains of *V. inaequalis* may differ in their sensitivity to fungicides. Some of the differences between isolates in Table 1 treated with the same fungicide appear to be greater than those expected from variation between different assays.

There was no indication that isolates from sprayed orchards were more resistant to fungicides than those from unsprayed orchards. The isolates were sensitive to a wider dosage range of sulphur than of other fungicides (Table 2). The widespread use of sulphur fungicides on apples in this area for many years may have caused this variation in the sensitivity of the isolates to sulphur. Where several isolates were from the same variety or the same orchard, there was no indication that the variety or orchard had any influence on the sensitivity of the isolates to the fungicides.

TABLE 2.—COMPARATIVE SENSITIVITY OF 50 ISOLATES of *V. inaequalis* TO VARIOUS FUNGICIDES

Fungicide	Initial conc. p.p.m.	Number of isolates completely inhibited at various dilutions					
		0	½	¼	⅛	1/16	1/32
Glyodin	10	50	38	15	0	0	0
Sulphur	10	50	48	45	26	1	0
Copper sulphate	20	35[1]	39	0	0	0	0
Captan	5	50	38	8	1	0	0
Dichlone	0.5	50	37	13	2	0	0
Phenyl mercury acetate	0.05	47	17	16	0	0	0

[1]Only 36 isolates were tested at this concentration of copper sulphate

Parry and Wood (11, 12) were unable to obtain strains of *V. inaequalis* resistant to captan but they suggested that, with the increasing use of fungicides more selective in their action than many of the older fungicides, resistant strains of fungi may appear more frequently. The results in this paper give no indication that this has occurred with *V. inaequalis*.

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PETAL BLIGHT, A NEW DISEASE OF THE CANNA 'PFITZER'S DWARF'[1]

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ABSTRACT

A severe petal blight was found affecting the canna 'Pfitzer's Dwarf' at Ottawa in 1959. The principal cause proved to be a pathogenic strain of an *Alternarai* of the *tenuis* type. Petal infection readily occurred from 5° to 30°C. but lesions developed most rapidly at 25°C. The fungus also infected wounded leaves of canna, but it caused little decay. *Myrothecium verrucaria* (Alb. & Schw.) Ditmar ex Fr., isolated once from a diseased petal, could infect petals but it caused less rapid decay than the *Alternaria*.

INTRODUCTION

The canna cultivar 'Pfitzer's Dwarf' with pink, red, yellow, and salmon flowers has recently been introduced as a promising ornamental. Forty of these plants were grown in a test garden at Ottawa in 1959. Eighty to eighty-five per cent of the plants became severely affected with a petal blight during a hot, humid period in late August. Plants of each flower colour were equally susceptible to the petal blight. The disease, of fungus origin, appeared to be new. Since its effects were devastating a study was initiated to determine the causal agent and some of the factors governing the development of the disease.

SYMPTOMS

Light-brown spots of various sizes were the first symptoms that developed on the infected petals. These spots enlarged and coalesced to involve large areas of each petal. The decayed areas were usually irregular in outline but occasionally spots in a row coalesced to form a streak. The affected petals finally became completely necrotic (Figure 1).

ETIOLOGY

A species of *Alternaria* sporulated profusely on diseased petals placed in a moist chamber. This fungus, identified as an *Alternaria* of the *tenuis* type, was also readily and repeatedly isolated from infected parts of petals plated on potato dextrose agar. In addition, *Myrothecium verrucaria* (Alb. & Schw.) Ditmar ex Fr. was isolated from one infected petal. Both fungi were cultured and tested for their pathogenicity to canna petals.

In pathogenicity tests, flowers and detached petals were inoculated with mycelium from single spore cultures of both fungi. In another trial, flowers were atomized with a water suspension of *Alternaria* spores. The detached petals were placed in moist chambers maintained at approximately 21°C. The plants, with inoculated flowers either covered with moistened plastic or left uncovered, were placed in a greenhouse maintained at 17°-22°C.

In the tests similar results were obtained with flowers and detached petals (Table 1). Both fungi readily infected the unwounded petals and caused lesions like those on the naturally infected flowers. Lesions caused by *Alternaria* on detached petals are shown in Figure 2. Infection by *Alternaria* was evident within 24 hours but infection caused by *M. verrucaria* was not evident until 48 hours. High humidity favoured infection by

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TABLE 1.—PATHOGENICITY TESTS WITH MYCELIUM OF *Alternaria* AND *Myrothecium verrucaria* ON FLOWERS[1] AND DETACHED PETALS OF CANNA

	Pathogen	Disease incidence[2]	Days from inoculation to lesion measurement	Average diameter of lesions (mm.)
Detached petals	*Alternaria*	54/54	5	16.6
	M. verrucaria	23/24	10	15.4
Flowers	*Alternaria*	30/33	3	7.6
	M. verrucaria	10/20	5	6.8

[1]The flowers were covered with plastic after inoculation
[2]*Numerator*—Number of infections
Denominator—Number of inoculations

mycelium of *Alternaria* as indicated by the high rate of infection on flowers covered with plastic. Only 20 per cent of such inoculations produced infections on flowers exposed to the atmosphere. Inoculations with *Alternaria* spores produced results similar to those with mycelium. *Alternaria* caused a more rapid decay of the tissues than *M. verrucaria* as shown by the average diameter of the lesions caused by the two fungi (Table 1). The results of these tests indicate that both fungi were pathogenic to the petals but that *Alternaria* was the more virulent.

Alternaria sp. has been reported as the cause of a leaf spot of *Canna generalis* Bailey (8) but its role as the primary pathogen was questioned. Therefore, attached leaves of canna 'Pfitzer's Dwarf' were inoculated with mycelium of the *Alternaria* from diseased petals to determine whether the fungus could cause a leaf spot. Leaves wounded with an inoculating needle prior to inoculation developed small brown lesions with yellow borders after 18 days at approximately 23 per cent of the inoculation points. Inoculations on unwounded leaves produced negative results. These results indicate that the isolate of *Alternaria* which readily caused a decay of the uninjured petals is only a weak wound pathogen on the leaves.

TEMPERATURE RELATIONS

The effect of temperature on the growth of the pathogenic strain of *Alternaria* on potato dextrose agar over a 1-week period was determined. The results are shown in Figure 3.

In another test the effect of temperature on infection and colonization of detached petals was studied. Six inoculations with mycelium were made on each of 12 petals placed in moist chambers. Inoculated and check petals were incubated at temperatures of 5°, 10°, 15°, 20°, 25°, and 30°C. The results obtained after a 4-day incubation period are given in Figure 4. Infection did not occur in 4 days at 5°C., but in 9 days a lesion had developed at each point of inoculation. At 30°C., only three lesions developed on one petal. Eight lesions developed at 25°C. and 12 at each of the other temperatures. Maximum diameter of the lesions was obtained at 25°C., indicating that it was the optimum temperature for development of the disease.

Physiological deterioration of the petals developed at the three higher temperatures. The symptoms of this condition expressed by the petals

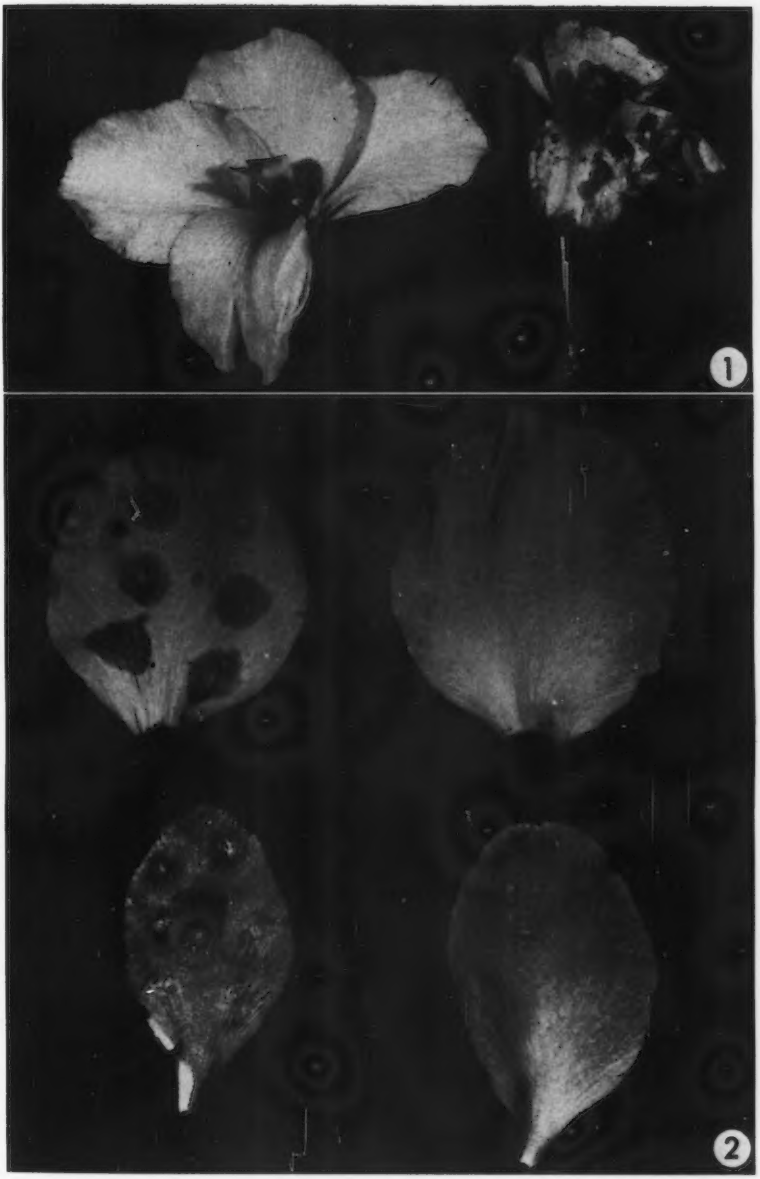

FIGURE 1. *Left*, a healthy pink flower of the canna 'Pfitzer's Dwarf'. *Right*, a pink flower affected with petal blight.

FIGURE 2. Petals showing lesions 4 days after inoculation with mycelium of *Alternaria*. The lesions on both petals were similar but those on the *upper pink* petal photographed more clearly than those on the *lower dark red* one.

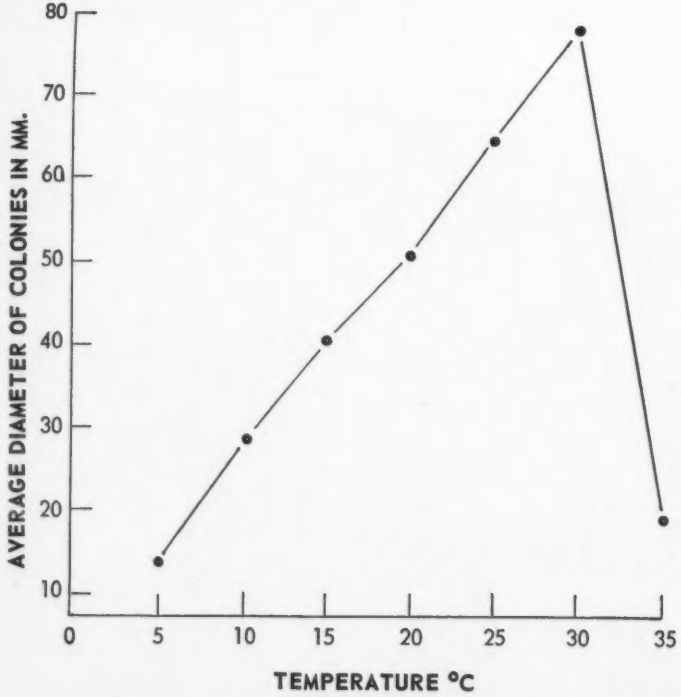

FIGURE 3. The average diameter of six colonies of the pathogenic strain of *Alternaria* of the *tenuis* type grown on potato dextrose agar for 1 week at seven temperatures.

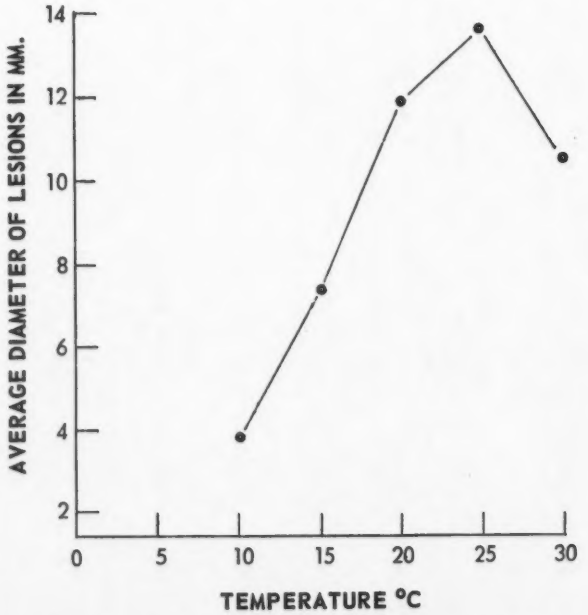

FIGURE 4. The effect of temperature on infection and colonization of canna petals by *Alternaria* as shown by the average diameter of the lesions 4 days after inoculation. In 4 days no infection had occurred at 5°C., but in 9 days lesions with an average diameter of 4.8 mm. had developed.

varied from a slight purplish discoloration of the edges at 20°C. to the development of a brown margin at 25°C. and finally to most of the petal turning brown at 30°C.

DISCUSSION

An *Alternaria* of the *tenuis* type and *Myrothecium verrucaria* were found in this study to be pathogenic to petals of the canna 'Pfitzer's Dwarf'. Neither fungus has apparently been reported as causing a disease of canna flowers. The results of the experiments reported here indicate that the *Alternaria* is the principal cause of the disease. Most strains of *Alternaria tenuis* auct sensu Wiltshire appear to be non-pathogenic (2). Nevertheless, both *A. tenuis* and *Alternaria* of the *tenuis* type have been reported as the cause of a number of diseases (1, 3, 4, 6, 7).

Although *M. verrucaria* caused infection of the petals, lesions developed much more slowly than those caused by *Alternaria*. *M. verrucaria* is generally regarded as a saprophyte but one of the cultures studied by Preston (5), isolated from potato haulms, grew readily on detached but living potato haulms and cucumber leaves. In the present study, *M. verrucaria* was isolated from only one diseased petal, which suggests that it was of relatively little importance in causing the high incidence of petal blight. On the other hand, *Alternaria* was the dominant fungus isolated from the diseased petals. The spores of this fungus may be disseminated by air currents and, under suitable environmental conditions, they could cause numerous infections resulting in a high incidence of disease.

In these studies decay of the petals by *Alternaria* was found to occur over a wide temperature range, indicating that temperature would not be a limiting factor in the development of petal blight. The disease is likely to be most severe under conditions of high humidity and moderately high temperature.

ACKNOWLEDGEMENTS

The writer wishes to express his thanks to J. W. Groves, of the Plant Research Institute, and E. G. Simmons, of the Pioneering Research Division, Q.M. Research and Development Centre, Natick, Mass., for confirming the identification of the species of *Alternaria;* and to S. J. Hughes for identifying the fungus *Myrothecium verrucaria.*

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THE ORGANIC ACIDS IN SILAGE AS DETERMINED BY GAS CHROMATOGRAPHY[1]

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ABSTRACT

A method of extracting organic acids from silage and of separating them in the esterified form by gas chromatography is described. The method was applied to a study of the volatile fatty acids and of lactic and succinic acids in alfalfa silage. Fresh alfalfa contained small amounts of lactic, acetic and succinic acids. The quantities of these acids increased during fermentation of the silage but succinic and lactic acids decreased at later stages of fermentation, while butyric acid became prevalent. Wilting of the alfalfa crop for 8 hours prior to ensiling decreased the rate of formation of all acids and prevented the decrease of lactic and succinic acids. Butyric and propionic acids were present in only small quantities. A more severe wilting treatment decreased the rate of formation of acids still further. Some acids not detected by other methods were found in trace amounts.

INTRODUCTION

During recent years the determination of organic acids has received the attention of numerous workers. Methods have been developed or adapted for the separation and identification of individual silage acids or groups of acids. Virtanen and Miettinen (18) and Duncan and Porteous (7) used paper chromatography to separate the lower volatile acids. Hirst and Ramstad (8) accomplished the same purpose with a silica gel column. Purves (14) modified the counter-current distribution technique of Sato and associates (16) for silage acids. Wiseman and Irvin (19) developed a celite column with the visibility advantages of an internal indicator that could separate formic, acetic, propionic, butyric, lactic and succinic acids. Gas chromatography as a means of separating volatile fatty acids was introduced in 1952, by James and Martin (9). The method, capable of resolving acids from formic through dodecanoic in a one-column analysis, was used for silage by Barnett and Duncan (5). With most of the methods, it was necessary to determine lactic acid as a separate entity by the colorimetric method of Barker and Summerson (2) as modified for silage by Barnett (3). A new approach to the determination of non-volatile acids was offered when James and Martin (10) separated and estimated the methyl esters of fatty acids by gas chromatography. The present work includes the description of a method developed for the determination of volatile fatty acids and other silage acids such as succinic and lactic acids by gas chromatography. The method was applied to study the formation of organic acids in alfalfa ensiled at three moisture levels. Laboratory silos were used in order to permit a better control of environmental conditions.

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MATERIALS AND METHODS

The silages were prepared from a third cutting of alfalfa in the pre-bloom stage in September 1958, at St. Paul, Minnesota. The forage was cut with a garden tractor. One-third was taken to the laboratory at once; the remainder was allowed to wilt in the field, one-third for 8 hours, and one-third for 24 hours, before being taken to the laboratory. The moisture contents of the resulting three fractions were 75.2, 62.7, and 55.3 per cent, respectively. They will be referred to hereafter as "high-", "medium-" and "low-moisture" treatments. In all cases the forage was transported to the laboratory in large plastic bags, chopped uniformly to half-inch lengths and ensiled within 2 hours after leaving the field. Six laboratory silos of a capacity of about 5 pounds were filled with material at each of the three moisture contents. Six 500-gram samples from each treatment were taken at filling time and stored in a freezer at −5°C., pending analysis.

The experiment was performed in laboratory silos equipped for the control of pressure and temperature (12). The silos were operated under anaerobic conditions at a temperature of 33.8° ± 0.5°C. A constant pressure of 12 inches of mercury was maintained on the silage throughout the storage period by means of a piston activated by manostat-controlled compressed air.

Two silos from each treatment were emptied after the following storage periods: 36 hours, 8 days and 240 days. The selection of the periods was based on the results of previous experiments. As measured by gas evolution, bacterial activity had reached a maximum about 36 hours after the crop had been ensiled and had practically ceased at the end of 8 days. The 240-day period was selected to provide a measure of the effect of the wilting treatment on the conservation of the resulting silage. Composite samples of silage were taken at unloading time and stored in a freezer in plastic bags.

Analytical Methods

Dry matter determinations: The per cent dry matter of initial forages and of silage was determined in triplicate by the toluene distillation procedure (1).

Total Acidity: The total acidity was obtained by potentiometric titration of the silage extracts after passage through a strongly acidic cation exchange resin to remove amino acids and cations from solution.

Extraction and purification of acids from silage and fresh forage: Ten-gram samples of silage or fresh forage were macerated in a Waring blender for 10 minutes with 100 millilitres deionized distilled water. The extracts were then heated to 100°C. under a reflux condenser, cooled to room temperature, filtered with slight suction through Whatman No. 40 filter paper, and the residues washed with three 25-millilitre portions of boiling water. Loss of volatile acids through the filter pump was minimized with a cold water trap. Filtrate and condensed liquids in the trap were pooled and preserved with 10 millilitres of a 5 per cent (W/V) solution of thymol in alcohol.

The extracts were purified by passage through a strongly acidic cation exchange resin (Amberlite IR-120, 16-50 mesh, in the free acid form) to remove amino acids and cations. The columns were rinsed with 100 millilitres of 40 per cent ethanol (V/V) to elute long chain monocarboxylic acids. The eluates were then passed through a weakly basic anion exchange resin (Amberlite IR-45, in the hydroxyl form) to retain the free acids and allow sugars and some of the pigments to pass through. The acids on the Amberlite IR-45 resin column were eluted with 100 millilitres of 0.5 N-NaOH and the excess NaOH removed by passage through another Amberlite IR-120 resin column. Recovery of standard acetic, propionic, lactic and succinic acids after passage through the columns ranged from 94 to 100 per cent of the theoretical value, whether the acids were run singly or in mixtures. The recovery of butyric acid was 91.3 ± 3.0 per cent.

Preparation of Acid Esters

The solutions containing acids were neutralized with excess dilute NaOH and concentrated by distillation under reduced pressure followed by drying over a steam bath to approximately 0.2 millilitre (4). Approximately 4 grams of anhydrous $KHSO_4$ were added to the cooled solution with 1 drop of phenol red indicator. The mixture was shaken until a dry pink powder was obtained following which the acids were extracted with six 4-millilitre portions of anhydrous ether. Ether was removed by fractional distillation and the acids were then esterified with an alcohol-free ethereal solution of diazomethane according to De Boer (6).

The esterified solutions were made to 25 millilitres with ether and a 5-millilitre aliquot removed for determination of the lower fatty acids. The remaining 20-millilitre portion was concentrated under reduced pressure at 0°C. and diluted to 5 millilitres with ether for the determination of esters of higher boiling point.

Gas Chromatography

A commercial gas chromatography apparatus (Beckman model GC-2) equipped with a thermal conductivity detector was used for analyses of esterified acid preparations from silages. The chromatographic column (Beckman Instruments Inc., column No. 17449) consisted of a 6-foot length of ¼-inch stainless steel tubing, filled with 15 grams of 42-60 mesh crushed C-22 Johns-Manville firebrick, coated with 8 millilitres of Dow-Corning Silicone Fluid type 550. A satisfactory resolution of the lower fatty acid esters was obtained with a column temperature of 100°C., a pressure of the helium carrier gas of 20 p.s.i. at the column inlet and a filament current at 250 milliamperes. A second sample of fatty acid esters was run at the same instrument conditions but at a column temperature of 190°C. for resolution of higher boiling esters. Samples were introduced into the column through a self-sealing serum cap by using a syringe type liquid sampler with a 1-inch hypodermic needle.

Identification of esterified acids was achieved by comparison of the retention time of esters of silage acids with the retention time of pure esters. A typical elution curve at a column temperature of 100°C. is shown in

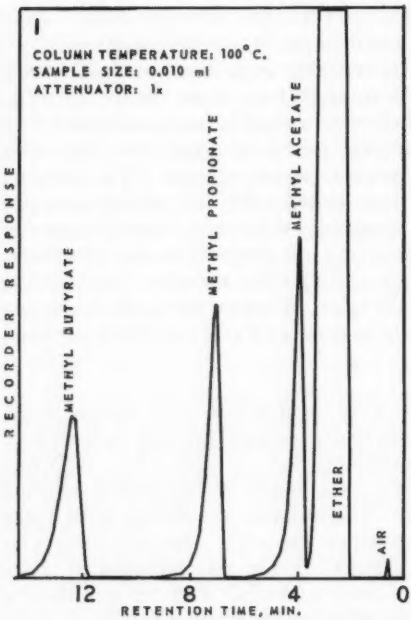

FIGURE 1. Chromatogram of a mixture of the methyl esters of acetic, propionic and butyric acids in ether.

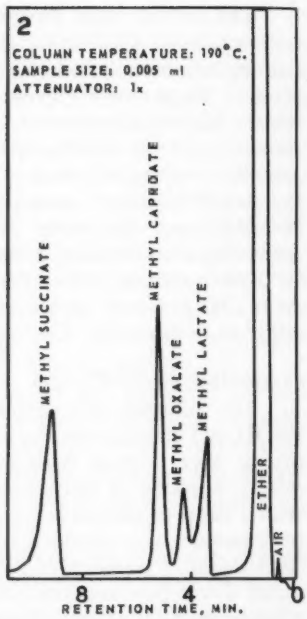

FIGURE 2. Chromatogram of a mixture of the methyl esters of lactic, oxalic, caproic and succinic acids in ether.

Figure 1 for esters prepared from pure fatty acids. The elution curve of esters of lactic, oxalic, caproic and succinic acids at 190°C. is shown in Figure 2.

Calibration curves for known esters were prepared as follows: volumes of 0.1 to 1.5 millilitres of each of the pure esters were diluted to 25 millilitres with anhydrous ether in volumetric flasks and 0.005-millilitre samples of the solutions were added to the column. A separate calibration curve was prepared for each acid. The concentration of the esters was determined by measuring the area under elution curves or alternatively the height of the peaks. When known amounts of acids were added to silage extracts and run through the entire procedure, the recoveries for acetic, propionic, butyric, lactic and succinic acids were respectively 79.5, 87.7, 88.2, 92.1 and 99.2 per cent on the average of three trials. Reproducibility as indicated by the results of duplicate determinations of a number of silage samples was within ± 5 per cent.

RESULTS AND DISCUSSION

Gas-liquid partition chromatography of the organic acids as the methyl esters proved to be a suitable method for the determination of organic acids in silage. In the purification procedure for silage extracts the only com-

pounds not removed by the resin columns were some of the pigments which did not interfere with the chromatographic separations obtained. The absence of contamination by other anions in the silage extract was also a desirable feature in the analytical procedure.

The scope of this study was restricted to the acids known to be of importance in silage, namely: lactic, butyric, propionic, acetic and succinic acids. However, a number of other peaks were present on the chromatograms which did not conform to the known standard curves. A number of these peaks were tentatively identified as isobutyric, isovaleric, valeric, isocaproic and malonic acids by the procedure of James and Martin (10)

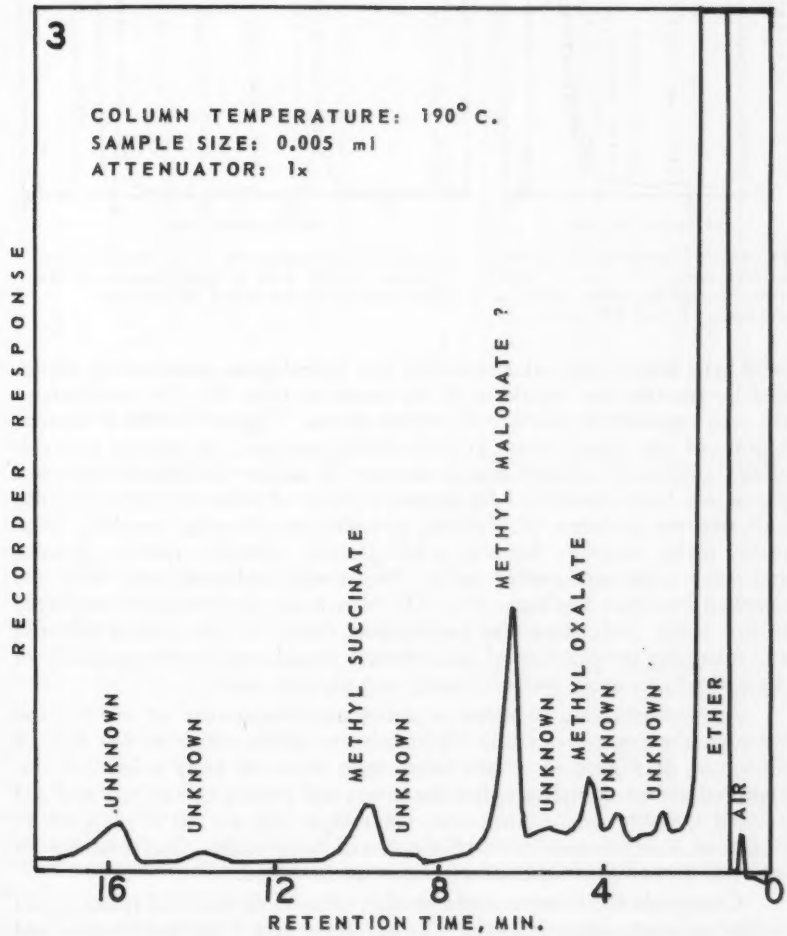

FIGURE 3. Chromatogram of the esterified acids in fresh unwilted alfalfa.

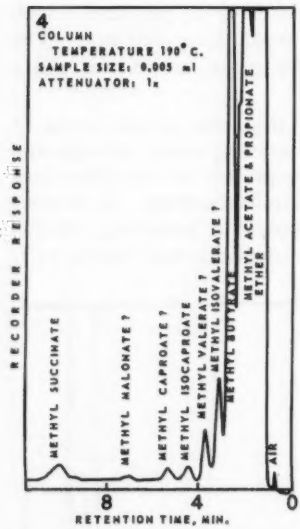

FIGURE 4. Chromatogram of the methyl esters of acids in high moisture alfalfa silage after a storage period of 240 days.

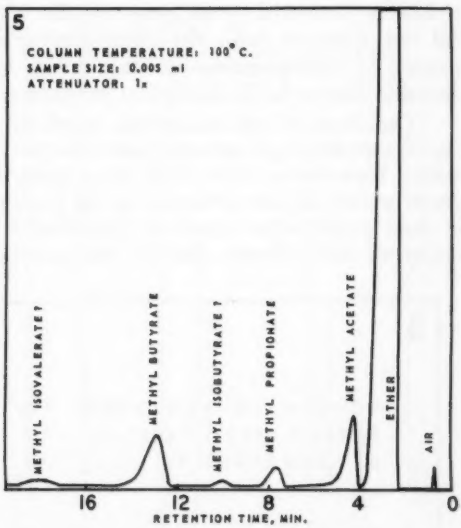

FIGURE 5. Chromatogram of the methyl esters of lower volatile acids in high moisture alfalfa silage after a storage period of 240 days.

who have shown that unknown acids in a homologous series can be identified by plotting the logarithm of the retention time of a few members of the series against the number of carbon atoms. Figure 3 shows a chromatogram of the organic acids in fresh alfalfa material. It may be seen that alfalfa contained a much higher number of acidic compounds than had previously been reported. An extensive study of acids in alfalfa (15) had indicated the presence of glyceric, pyrrolidonecarboxylic, succinic, citramalic, malic, malonic, fumaric, α-ketoglutaric, shikimic, quinic, glycolic, and some other unidentified acids. Oxalic acid and citric acid were also detected by other workers (13). Of these acids the procedure employed in this study (extraction and purification, nature of the column material, and operating temperature of the column) would restrict the possibility of detection to succinic, malonic, oxalic and glycolic acids.

A chromatogram obtained at a column temperature of 190°C., and showing the organic acids in high moisture alfalfa silage at 240 days, is illustrated in Figure 4. Peaks other than those of fatty acids had disappeared almost completely, but there was still a trace of malonic acid and a small quantity of succinic acid. A sample run at 100°C. (Figure 5) produced a better resolution of the lower fatty acids. The presence of iso-acids as well as straight-chain acids may be noted.

Changes in the relative concentration of succinic acid and malonic acid in low moisture silage are shown in Figures 6 and 7 for the 36-hour and the 240-day fermentation periods respectively. It may be of significance

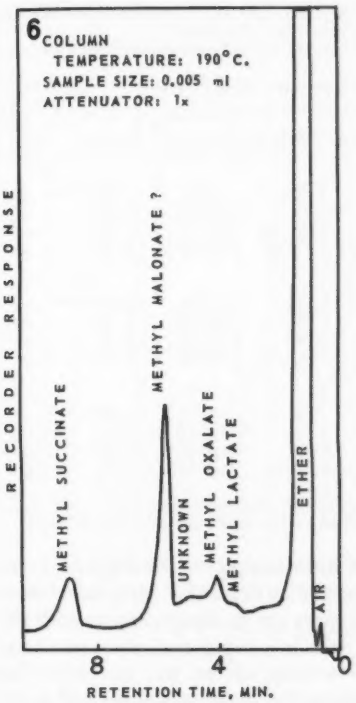

FIGURE 6. Chromatogram of the methyl esters of acids in low moisture silage after a storage period of 36 hours.

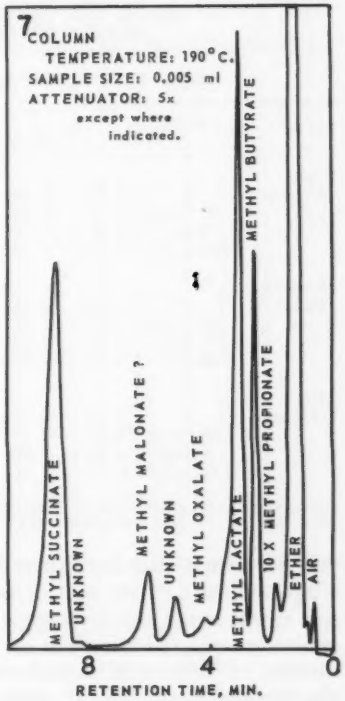

FIGURE 7. Chromatogram of the methyl esters of acids in low moisture alfalfa silage after a storage period of 240 days.

that, while succinic acid increased during fermentation, malonic acid decreased correspondingly. Oxalic acid appear in small and constant amounts in all silages except in the 240-day high-moisture silage.

The organic acid content of high-, medium- and low-moisture alfalfa silages at different periods after ensiling is shown in Table 1, together with the pH value of the corresponding silages. The total acidity showed a tendency to decrease during wilting in the field, but increased during fermentation. The increase was more noticeable in silage at high moisture level in the 8-day sample, but there was a subsequent decrease in the 240-day sample, probably as a result of the disappearance of lactic acid. Corresponding figures for total acidity are difficult to find in the literature since the term was most often used to represent the sum of volatile acids and of lactic acid, without taking into account other non-volatile acids.

There was always some lactic acid present in the initial crop. The concentration increased rapidly in the high-moisture silage but at a slower rate in wilted silage. Noteworthy is the fact that lactic acid accounted for

TABLE I.—THE ORGANIC ACID CONTENT OF HIGH-, MEDIUM- AND LOW-MOISTURE ALFALFA SILAGE AT DIFFERENT PERIODS AFTER ENSILING

Treatment	Time after ensiling	pH	Acid content, per cent of dry matter					Total acidity: me./gm. D.M.
			Acetic	Propionic	Butyric	Succinic	Lactic	
High-moisture	0	5.75	0.56	0	0	0.17	0.08	1.89
	36 hours	5.58	1.48	0	0	0.62	4.90	1.84
	8 days	5.64	2.74	0.16	0.68	1.14	6.73	2.62
	240 days	5.56	3.52	1.38	4.94	0.37	0.00	2.23
Medium-moisture	0	5.64	0.35	0	0	0.13	0.02	1.68
	36 hours	5.94	1.04	0	0	0.21	0.20	1.80
	8 days	5.74	1.18	0	0	1.06	4.14	1.95
	240 days	4.98	1.90	0.23	1.74	1.11	7.52	2.21
Low-moisture	0	5.75	0.77	0	0	0.12	0.09	1.42
	36 hours[1]	5.95	0.49	0	0	0.18	0.16	1.68
	8 days	5.96	0.60	0	0	0.35	0.30	1.51
	240 days	4.92	1.23	0.15	0.74	0.76	5.53	1.81

[1]One of the two replicates was discarded as a consequence of air contamination during the storage period.

4.90 per cent of the dry matter in the 36-hour sample of the high moisture silage and for 6.73 per cent of the dry matter in the 8-day sample, while it was absent in the 240-day sample. The results are in disagreement with the generally accepted theory according to which a rapid and abundant production of lactic acid should ensure a lowering of the pH necessary for the production of "stable" silage. In this case a high production of lactic acid did not lower the pH appreciably and did not increase the total acidity in the 36-hour sample. This would suggest that the acids originally present in the fresh forage were destroyed as fast as sugars were fermented, with the result that the acidity was maintained at about the same level.

The data in Table 1, using production of acetic, propionic, butyric, succinic and lactic acids as an index of bacterial activity, are consistent with the report of Stirling (17), who has shown that wilting causes a delay in bacterial multiplication. The content of lactic acid in the final silages is in general agreement with corresponding values reported by Murdoch (11) for silages of different dry-matter contents, except that much higher values are reported in this work. It must be remembered, however, that air-tight laboratory silos were used in this study, thereby excluding possible mould growth and oxidation from air contamination which may have a decided effect on the various acid components. Using the criteria of high lactic acid and low butyric acid content as a basis for evaluating silage quality, it is apparent from Table 1 that wilting alfalfa to medium- or low-moisture levels resulted in the production of "good" silage with a relatively high concentration of lactic acid and little butyric acid over an 8-month conservation period. Also it is apparent from Table 1 that the pattern of acid appearance and/or disappearance is remarkably consistent among the three silages.

TABLE 2.—THE ORGANIC ACIDS IN HIGH-, MEDIUM- AND LOW-MOISTURE ALFALFA SILAGE AS MILLIEQUIVALENT PER CENT OF THE TOTAL ACIDITY

Treatment	Time after ensiling	Meq. per cent of total acidity					
		Acetic	Propionic	Butyric	Succinic	Lactic	Other
High-moisture	0	4.9	0	0	1.5	0.5	93.1
	36 hours	13.4	0	0	5.9	29.4	51.4
	8 days	17.4	0.8	3.0	7.4	28.6	42.8
	240 days	26.3	8.4	25.2	2.8	0.0	37.4
Medium-moisture	0	3.5	0	0	1.3	0.1	95.1
	36 hours	9.7	0	0	2.0	1.2	87.1
	8 days	10.2	0	0	9.2	23.5	57.6
	240 days	14.3	1.4	8.9	8.5	37.8	29.2
Low-moisture	0	9.0	0	0	1.4	0.7	88.9
	36 hours[1]	5.1	0	0	1.8	1.1	92.1
	8 days	6.6	0	0	3.9	2.2	87.4
	240 days	11.4	1.2	4.7	7.1	34.0	41.7

[1]One of the two replicates was discarded as a consequence of air contamination during the storage period.

The relative importance of the acids with regard to the total acidity was calculated from the following formula:

$$\text{me\%} = \frac{\text{Wt. of acid per gram of dry matter x 100}}{\text{me. wt. of acid x me. total acid per gm. D.M.}}$$

The results of the calculations are shown in Table 2. It is apparent that acetic, propionic, butyric, lactic and succinic acids accounted for only a small proportion of the total acidity. This was especially the case for the initial samples. As fermentation progressed, acetic, butyric and lactic acids accounted for an increasingly higher percentage of the total acidity. It is apparent also that the increase in those acids was not followed by a corresponding increase in the total acidity, suggesting that a proportion of the original acids was either destroyed or used up in the formation of fatty acids and of lactic acid. The fate of the so-called plant acids during fermentation would deserve further investigation.

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RESPONSE OF LITTLE CHERRY-INFECTED CHERRY TREES TO ZINC TREATMENTS[1]

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ABSTRACT

In the Creston Valley of British Columbia, mature Lambert cherry trees displaying severe symptoms of the virus disease little cherry were treated with various zinc carriers applied to the soil, to the dormant wood, or to the foliage. The trees had not shown the chlorotic symptoms typical of zinc deficiency but had produced fruits and leaves that were smaller than might be expected from the virus infection alone.

The zinc treatments invariably resulted in improvements in leaf size, and especially in fruit quality. The best zinc treatment gave increases of 59 per cent in fruit size and 77 per cent in soluble solids content. The most marked responses were obtained with zinc sulphate applied as a dormant spray or with ZnEDTA chelate applied to the soil. Foliar sprays were the least effective. The quantity of zinc present in the leaves collected from treated plots showed no association with observed fruit responses. Leaf analyses demonstrated that a mild zinc deficiency condition existed. These trials indicate that the quality of fruits on trees infected with little cherry virus may be adversely affected by zinc deficiency, in the absence of recognizable zinc deficiency symptoms.

INTRODUCTION

Zinc-deficient sweet cherry trees exhibit the "little leaf", "rosette", and "dieback" symptoms that characterize the deficiency in many kinds of fruit trees. The leaves are chlorotic in the interveinal areas, twisted and distorted, and generally reduced in size. In addition to these commonly observed symptoms, the fruits may be smaller than normal (5), and may be abnormal in colour at maturity. When the deficiency is only mild, the more obvious symptoms may be lacking, and the condition may, therefore, escape detection.

The virus disease little cherry does not cause obvious leaf symptoms in most commercial sweet cherry varieties. Fruit symptoms are not apparent until picking time. At this stage, many or all of the fruits on infested trees are half or less than half normal size and retain the light red colour of immature cherries (4). In the Kootenay Valley of British Columbia, virtually all cherry trees are infected with the little cherry disease. In Creston Valley orchards there are wide variations in the degree to which the little cherry disease reduces fruit size. The differences are most apparent from orchard to orchard, but sometimes pertain to parts of a single large planting. An infected tree may produce fruits of marketable quality in occasional years, but, more often, trees that are severely affected consistently yield poor fruit. This has suggested that another factor is intensifying the effect of the virus in severely affected orchards.

Further, mild symptoms of zinc deficiency had been observed and confirmed in a few apple plantings in the area. Although distinctive symptoms had not been seen in cherry, it is quite as susceptible to zinc deficiency as apple (3). For this reason, and because of the similarity of

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TABLE 1.—RATE AND TIMING OF ZINC TREATMENTS APPLIED TO CHERRY TREES IN 1958 AND 1959 TRIALS

Material	Rate	Time of application
1. ZnEDTA[1] soil application	½ lb./tree	When trees fully dormant
2. ZnEDTA soil application	1 lb./tree	When trees fully dormant
3. $ZnSO_4$ spray[2]	40 lb./100 gal.	Before bud-break
4. $ZnSO_4$ spray followed by dormant oil spray	40 lb./100 gal. 4 gal./100 gal.	Before bud-break
5. $ZnSO_4$ spray	32 lb./100 gal.	When buds open, but before blossoms separated from cluster
6. $ZnSO_4$ spray followed by Plyac[3] spray	32 lb./100 gal. 4 oz./100 gal.	When buds open, but before blossoms separated from cluster
7. $ZnSO_4$ plus lime spray	6 lb.+6 lb./100 gal.	When leaves fully expanded
8. ZnEDTA spray	2 lb./100 gal.	When leaves fully expanded
9. Check		

[1]ZnEDTA disodium zinc ethylenediamine tetra-acetate dihydrate
[2]$ZnSO_4$—containing 36% actual zinc
[3]Plyac—a non-ionic polyethylene surfactant which protects deposits against weathering

fruit symptoms resulting from the two disorders, tests were conducted to determine whether zinc deficiency was increasing the severity of little cherry symptoms.

Although soil applications of ZnEDTA have proven effective in treating zinc-deficient sweet cherry trees in Washington (2), cherry does not respond readily to standard methods of treatment (2, 6). Various zinc carriers and methods of application were, therefore, employed in attempts to enhance absorption of the element.

MATERIALS AND METHODS

All tests were conducted in an orchard planted with 20-year-old Lambert cherry trees that had exhibited moderate to severe little cherry symptoms for over 5 years. In preliminary trials, conducted in 1956 and 1957, zinc sulphate was applied at rates of 20-35 lb./100 gal. to the dormant trees with and without subsequent foliar sprays of zinc oxide at 1 lb./100 gal. Delayed dormant applications of zinc sulphate were also made to determine the latest stage in the development of the buds at which zinc sulphate could be safely applied. In 1958 unreplicated plots of 5 to 9 trees each, were treated with zinc compounds as outlined in Table 1. These treatments were repeated on the same trees in 1959.

Fruits were gathered from all treated and check plots at harvest time in 1958 and 1959 for weight and soluble solids determinations. In each plot, 100 fruits were sampled for weight measurements and 25 fruits for soluble solids determination. Soluble solids were determined on the expressed juice of each fruit individually by means of a refractometer. For ease of comparison, an index figure for fruit quality was calculated by multiplying the average weight by the average soluble solids content of the fruits from each plot.

Median shoot leaves were gathered from the plots on June 11, 1958, for analysis of zinc content. The leaves were first washed in tap-water, then in a solution of Dreft detergent, and finally in distilled water, in order to remove surface residues prior to drying and preparation for analysis. The zinc content was determined by the A.O.A.C. single colour dithizone method (1).

RESULTS

The responses to 1956 and 1957 treatments (zinc sulphate dormant spray – 25/100, followed by summer sprays of zinc oxide – 1/100) were inconsistent, although encouraging. The inconsistencies are attributable in part to the limited range of zinc carriers tested, and especially to the moderate injury suffered by many of the test trees during severe cold weather in early November, 1955. The experience in these two seasons was especially useful in indicating tree injury hazards that can be involved in foliar application of zinc compounds. These are described and discussed later in this paper.

The results of tests made in 1958 and 1959 with different zinc carriers and modes of application are reported in Table 2. Fruits from trees that received these zinc treatments were noticeably larger in size and contained

TABLE 2.—LEAF ZINC CONTENT AND FRUIT QUALITY OF CHERRIES AS AFFECTED BY VARIOUS ZINC TREATMENTS

Treatment (*see Table 1*)	1958				1959		
	Fruit weight, grams	S.S.C.[1] %	Index[2]	Leaf zinc p.p.m.	Fruit weight, grams	S.S.C.[1] %	Index[2]
Check	4.1	9.1	37	18.8	4.5	13.3	60
Dormant zinc	6.5	16.1	105	16.2	6.2	15.3	95
Dormant zinc+oil	6.5	14.1	92	19.9	5.7	14.0	70
ZnEDTA half-pound	6.0	14.9	89	18.9	6.1	14.2	86
ZnEDTA 1 pound	6.5	13.4	87	23.7	7.0	17.7	123
Delayed dormant	6.2	12.8	79	44.4	6.3	14.4	90
Delayed dorm Plyac	6.5	14.4	94	34.9	6.7	16.4	110
ZnEDTA 1 pound 1958 spray 2 pound 1959 spray	5.0	12.1	61	36.0	5.2	13.7	71
$ZnSO_4$ +lime	5.9	13.4	79	28.9	5.3	14.5	77

[1]Soluble Solids Content
[2]Fruit weight x SSC

LSD at .05—7.8

a greater percentage of soluble solids in the 2 years of the trial. In no case were the symptoms of little cherry disease completely masked, but a much larger percentage of the fruits from treated trees met market standards.

Fruits averaging less than 6 grams in weight and less than 14 per cent in soluble solids content are not acceptable to the fresh fruit trade. In both years, fruits from check plots failed to meet either of these standards. On the other hand, fruits sampled from zinc-treated plots met market requirements (except those that received zinc as a foliar spray only, and the plot that received dormant zinc sulphate spray followed by a dormant oil spray in 1959).

On the basis of fruit size and soluble solids content, trees given a dormant zinc sulphate spray at 40 lb./100 gal. in 1958 showed the greatest improvement, followed by trees that received a delayed dormant zinc sulphate spray and Plyac. In 1959 the trees that had received 1 pound of ZnEDTA chelate in 2 successive years as a soil application produced the best fruits followed by the delayed dormant zinc sulphate-Plyac treatment.

The most promising treatment in 1958 (dormant zinc sulphate) showed a 59 per cent increase in fruit size and a 77 per cent increase in soluble solids content over check fruits; in 1959 the increases were 55 per cent and 33 per cent respectively in fruits from the best plot (ZnEDTA 1 lb./tree). Foliar treatments in both years proved inferior to the other methods of treatment tested. Nevertheless, response was demonstrated in all of the zinc-treated plots in both years.

Injury was sustained in all plots that received the delayed dormant sprays of zinc sulphate in 1957. The injury varied from slight marginal leaf burn to complete killing of leaf buds. Blossoms that had passed the cluster-bud stage when sprayed were partially or completely killed. Many of the fruits that developed from the injured blossoms abscissed during the June drop. As expected, the injury was most severe in the plot that was sprayed latest in the season (blossoms 0-50 per cent of full bloom). By picking time, this early season leaf injury was barely visible in the sprayed trees because of their response to the zinc treatments. This response was most evident in the effects on leaf size and colour. By the end of the growing season the density of the foliage was comparable in sprayed and unsprayed plots. Where blossom injury occurred, the trees carried a reduced crop, but the resulting fruits were appreciably larger and of better quality than those from the check trees. It is important to note that hand thinning of blossoms does not increase the size of fruits on little cherry-infected trees.

In contrast to the results obtained in the 1957 trials, no injury occurred in any of the zinc-treated plots in 1958 or 1959. Cool and rainy weather conditions prevailed following the delayed dormant zinc sulphate applications in 1957, whereas in 1958 and 1959 the weather was warm and dry following application. Experience has shown that foliar applied zinc compounds are likely to injure tissues if cool wet weather follows their application.

DISCUSSION

The values reported for the zinc content of mid-shoot leaves gathered from the test plots on June 11, 1958 (Table 2) are suggestive of a mild zinc deficiency and lie within the range of values for zinc-deficient sweet cherries found by Benson *et al.* (2) in Washington State. Differences in the leaf content of zinc are significant only in the case of the four plots which were treated with delayed dormant or with foliar sprays of zinc compounds. The foliage of trees in these plots would be expected to carry relatively large amounts of zinc as a surface deposit. Though the leaves were thoroughly washed prior to analysis, it is questionable whether such residues can be entirely removed from the leaf surface by any washing procedure, and it is, therefore, likely that a proportion of the zinc found was present as surface contamination.

The zinc content of tree tissues often does not correlate well with the severity of symptoms. Benson and co-workers (2) cited figures for zinc-deficient apple trees which showed that extremely chlorotic leaves were higher in zinc content than normal-appearing leaves from the same tree. Zinc deficiency reduces leaf size, and they explain that, had the amount of zinc been expressed on a leaf area rather than a leaf weight basis, the more chlorotic tissues would have shown relatively lower amounts of zinc. This is probably true in the present case as well, since improvements in fruit size were accompanied by increases in leaf size. It will be observed that leaves sampled from the plot that showed the greatest response to the zinc treatment in 1958 (dormant zinc sulphate) actually contained the lowest concentration of zinc. Further, it must be remembered that the test trees were afflicted with two disorders, zinc deficiency and little cherry. With both disorders, the severity of symptoms varies within and between trees from year to year. Under these circumstances the zinc content of tissues might vary in an unaccountable fashion. Under the conditions of these tests it is not possible to determine whether the response in the cherries was due to a mild zinc deficiency alone or to added zinc *per se* on the virus condition.

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FLOWER, POD AND SEED DEVELOPMENT RELATIVE TO THE TIMING OF THE SEED HARVEST OF VIKING BIRDSFOOT TREFOIL *(Lotus corniculatus)*[1]

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ABSTRACT

Investigations into pod and seed development and flowering habit relative to the timing of the seed harvest of birdsfoot trefoil were conducted from 1953 to 1956 at Ithaca, New York.

The process of pod and seed development was divided into three stages: *pod elongation, seed development* and *seed maturation.* Only the last stage was of importance in determining the optimum time for seed harvest. Indicators of this stage were the succession of pod colours: light green, green-white and golden brown. Since these colours indicate changes in seed viability and pod moisture content they were the basis of determining when an individual pod should be harvested. While light green pods contained seed which was high in germination percentage, it was not until pods turned a green-white that seed attained its maximum weight. Pods of this colour were high in moisture and did not dehisce. It was concluded, therefore, that a pod should be harvested when it was green-white in colour.

Seed developed in the lowest or oldest three umbels accounted for approximately 92 per cent of the potential yield. Any increase over this figure brought about by a delay in harvest which would permit the younger umbels to reach maturity would be overwhelmingly offset by seed loss resulting from deshicence of the older umbels.

The optimum time of seed harvest may be determined by count or estimate of seed pod maturity. When 60 per cent of the pods are light green to white-green in colour the crop may be considered to be in the early stages of ripening. When 85 per cent of the pods are golden brown a large number will readily dehisce under conditions of low humidity. It is concluded, therefore, that birdsfoot trefoil seed harvest should begin when 70 to 80 per cent of the pods are mature.

INTRODUCTION

The level of seed production of birdsfoot trefoil varies from location to location and year to year. Growers have reported yields as high as 400 pounds per acre on some occasions and as low as 50 pounds on others. One of the barriers to obtaining high seed yields is the lack of knowledge of the optimum harvest time for this crop. The exact time to begin the harvest is difficult to gauge because of the flowering habit of this species which results in the presence of mature pods, immature pods and flowers at the same time. A delay in the harvest of the mature pods will result in seed loss due to the dehiscence of these pods. On the other hand, harvesting so as to retain these first developed mature pods means that seed from later developing pods is immature and thus the full potential of the seed crop may not be realized.

To help clarify this problem, Macdonald (3) in 1940 and 1941, and Anderson (1) in 1952 and 1953, investigated the development of pods and seeds of this crop. Both workers found that pods changed colour during the process of development and that these changes in pod colour were

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related to seed development and quality. Thus indicators of seed quality were established. These workers agreed that dark green pods contained immature seed, that cream coloured or green-white pods contained seed of high germination ability and that only brown pods contained fully mature seed. They were able to predict when an individual pod should be harvested. On the basis that brown pods contained mature seed, recommendations were made (2, 4, 7) for harvesting seed when the pods were golden brown in colour.

Since all pods do not mature at the same time, these recommendations did not completely solve the problem of when to begin the harvest, as no method was suggested for the determination of when the crop was at the correct stage. Further complicating the picture was the finding of Metcalfe *et al.* (5) and Macdonald (3) that brown pods tend to dehisce more readily under conditions of low humidity than do less mature pods.

A part of Anderson's research was devoted to a study of the change in the ratio of flowers, immature pods and mature pods as the season progressed. He recorded the proportion of mature pods throughout the development and maturation of the seed crop. A method was suggested for the determination of the time for beginning seed harvest. He suggested that harvesting should commence earlier than had been previously suggested (2, 4, 7). This was based upon the conclusion that light green pods contained viable seed which was physiologically mature. He therefore recommended that, to reduce seed loss from the shattering of very mature pods, a field of trefoil should be harvested when the maximum number of pods was between a light green and a light brown colour. However, harvesting at this stage may not allow for the full potential of the seed crop to be realized as flowers and immature pods which are incapable of producing viable seed are harvested along with the more mature pods.

The potential seed yield of birdsfoot trefoil is governed by the indeterminate flowering habit of this crop. No data are available to indicate the influence of this flowering habit on the total amount of mature seed available for harvest. Nor have the relationships between this potential, the timing of seed harvest, and the development of individual pods been established.

This report describes investigations which were conducted with Viking birdsfoot trefoil from 1953 to 1956 at Ithaca, New York. The objectives of these studies were to examine the development of individual pods, to determine the influence of flowering habit on the potential seed yield and the relationship of these to the best stage of maturity for seed harvest.

MATERIALS AND METHODS

To study the effect of advancing maturity on pod and seed development, investigations were carried out during 1955 and 1956 in a Viking space-plant nursery. Individual flowers in this nursery were tagged after they had been pollinated by bees. Subsequently, every 2 days, 20 pods arising from the tagged flowers were harvested at random. The length and diameter of the pods were measured and the colour of seeds and pods

recorded. Per cent dry matter of pods and seeds was determined. Beginning on the 12th day after pollination, and at subsequent 2-day intervals, two samples each containing 100 seeds were air-dried, then weighed and placed for 10 days in a germination cabinet at 70°F. to determine the percentage of total live seed (*hard* plus *soft*). Data on pod length, 1000-seed weight and total live seed were analysed according to a randomized complete block design.

To provide information on the flowering habit of this legume and to determine its relationship to seed harvest, four 10-plant samples were harvested from each of five population densities of 1 plant per 4, 9, 16, 36 and 144 square inches. The samples were harvested when seeds pods from the lowest inflorescence on a stem were light golden brown in colour. Pods were individually harvested from successive umbels. The number of pods per umbel and seeds per pod were counted. Two samples of 100 seeds each were used to determine seed weight and germination percentage.

Studies to determine the proper time for seed harvest were conducted in 1954 and 1955. Each year seed plots were harvested at four maturity gradients, based upon the proportion of mature to immature pods. The proportion of mature pods was determined by choosing at random two samples of 100 pods and separating the pods into the colours: green purple, light green, green-white and light golden brown. Pods of the last three colours were considered as mature. Harvested material was bagged, air-dried and threshed. Seed from each maturity gradient was subdivided into three seed classes; *round*, *shrunken* and *shrivelled*. Seed weight and germination were determined on two samples of 100 seeds from each class. The data were analysed as a randomized complete block design. Duncan's multiple range test was used to determine differences among means.

RESULTS AND DISCUSSION

Pod and Seed Development

In both years, 1955 and 1956, the developing pods of Viking underwent colour changes from a dark green or dark green-purple through various shades of green to a green-white and then to golden brown (Figure 1). Similarly, seed colour changed from a dark green through olive-green to a brown. Although there were continuous changes in the appearance and the viability of the seed, pod and seed development were characterized by three distinct physiological stages: *pod elongation*, *seed development* and *seed maturation*. Each was delineated by morphological or physiological characteristics which indicated relatively distinct phases of the pod and seed development process.

The pod elongation stage was most strongly characterized by increases in pod length and was considered completed when pods had reached their maximum length. Seed was very immature during this stage. Although the seed gradually increased in size it could not be separated easily from the pod until the pod had reached its maximum length. After air drying these seeds turned black, shrivelled and did not germinate. Throughout this stage the pods were dark green or dark green-purple in colour, small in diameter, and high in moisture per cent (Table 1 and Figure 1).

TABLE 1.—PERCENTAGE GERMINATION OF VIKING BIRDSFOOT TREFOIL SEED HARVESTED AT 2-DAY INTERVALS DURING DEVELOPMENT. 1955 AND 1956 SEASONS.

	Days after pollination																		
	12	14	16	18	20	22	24	26	28	30	32	34	36	38	40	42	44	46	48
1955																			
Stage	Pod elongation		Seed development		Seed maturation														
Viable[1]	0.0	0.5	65.0	85.5	88.5	87.5	95.0	97.0											
Non-viable[2]	100.0	98.0	34.5	8.0	8.0	6.5	4.5	3.0											
Live seed[3]	0.0	1.5	65.5	92.0	92.0	93.5	95.5	97.0*											
1956																			
Stage		Seed development									Seed maturation								
Viable[1]	0.0	0.0	0.5	0.0	0.5	2.0	6.5	7.0	22.5	45.5	74.3	80.0	73.0	71.2	76.5	86.5	78.5	63.0	78.0
Non-viable[2]	100.0	100.0	99.5	100.0	99.5	92.5	89.0	83.0	65.0	44.7	21.1	15.0	20.3	27.2	21.0	11.5	19.0	30.0	20.0
Live seed[3]	0.0	0.0	0.5	0.0	0.5	7.5	11.0	17.0	35.0	55.3	78.9	85.0	79.7	72.8	79.0	88.5	81.0	70.0	80.0*

* Duncan's multiple range test p = .05
[1] Viable — seed germinates only after scarification
[2] Incapable of germinating after air drying
[3] Live seed = total live seed (including hard seed)

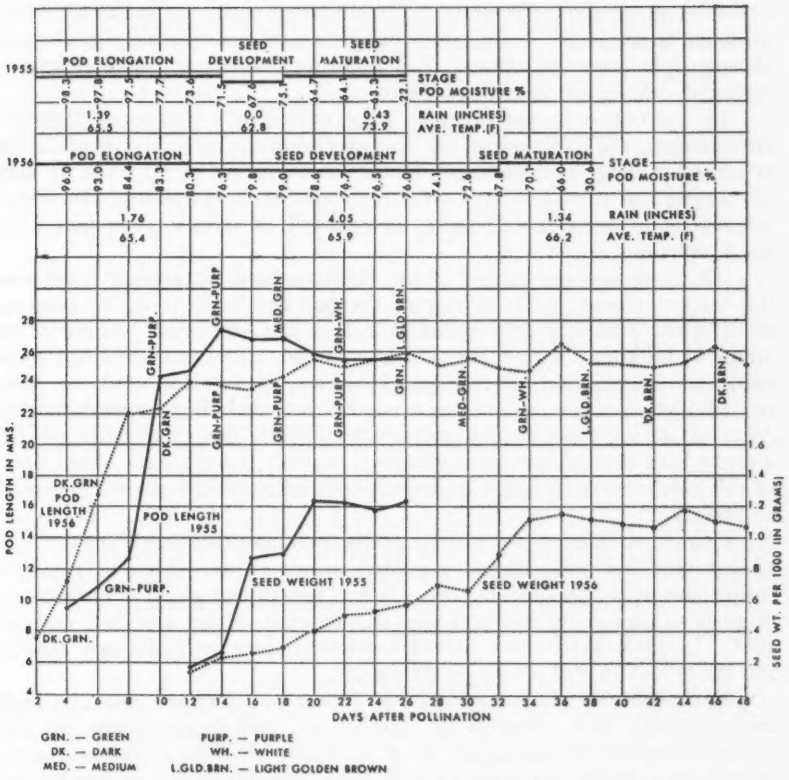

FIGURE 1. Development of pods and seeds of Viking birdsfoot trefoil during 1955 and 1956.

Little difference was noted in the time required for the completion of this stage between the 2 years (12 days in 1956 and 14 days in 1955). The average temperature was similar in both years but 0.35 inches more rain fell during this period of pod development in 1956 than in 1955.

The characteristic features of the seed development stage were marked increases in seed size and in germination percentage after air drying (Table 1 and Figure 1). Whereas at the beginning of this stage practically no seeds were capable of germinating, at the end just as many would germinate as would do so 2 weeks later. Although embryo size had reached its maximum and viability was high, food materials had not yet replaced the water in the cells of the seed and hence on drying the seeds were small, shrivelled and dark. Of the seeds viable at the end of the period practically all had acquired a hard seed coat, as shown by the fact that they did not germinate without scarification.

Coincident with the increase in seed size came an increase in the diameter of the pods. In this stage the latter lost their purple pigment and became light green in colour. Pod moisture content remained relatively high and dehiscence was not observed.

In contrast with the previous stage, which was influenced little by environment, the duration of the seed development stage was profoundly affected by climatic conditions. In the drier year of 1955, when no rain fell during this period, 4 days were required for completion of this stage, whereas in 1956, when 4.05 inches of rain fell, 20 days were required for its completion.

The conspicuous feature of the third or seed maturation stage was the marked change in the colour of the pod and in its moisture content. At first the pod was light green in colour and sufficiently succulent to inhibit dehiscence (Figure 1). During this phase the seeds attained their maximum germination percentage. However, it was not until the pods turned a green-white colour that seeds reached their highest seed weight. After air drying these seeds were rounded, slightly dented or flattened and did not have the characteristic glossiness of fully mature seed. Seed pods of this colour were still high in moisture percentage and were not observed to dehisce.

Coincident with a sharp drop in the pod moisture level, from 65 to 25 per cent, the pods turned a golden brown colour (Figure 1). Seed from these pods was fully mature. It was brown and glossy. Dehiscence of pods of this moisture level was encountered during the dry year of 1955. In 1956 under more humid conditions these pods did not dehisce and the colour turned progressively darker.

Environmental conditions did not appear to affect the rate at which seed matured. For, although the rainfall was 0.91 inches lower and the average temperature was higher by 8°F. during the seed maturation stage in 1955 than during the same stage in 1956, pods reached a golden brown colour in 8 days in 1955 and 6 days in 1956.

The significance of the above results can be understood more readily if viewed from the position of a seed grower who, by watching the development of a single pod, seeks to determine the optimum time for seed harvest. Seed harvested in the "Pod Elongation" or in the "Seed Development" stages can make no useful contribution to the seed yield except possibly at the end of the latter stage. Only when the pod is passing through the "Seed Maturation" stage has it the potential for making a significant contribution of good seed. All phases of the latter stage do not always provide equally for the production of good seed. Too early harvesting in this stage results in the production of seed which, although fully mature physiologically, will not be as plump or as well stocked with stored food as is desirable. Harvesting near the end of the period, on the other hand, greatly increases the danger of seed loss by dehiscence. It is suggested then that the proper compromise is to harvest just prior to the brown pod stage. At this time the seeds have reached their full germination power, food storage is complete and the danger of pod dehiscence is negligible.

TABLE 2.—THE NUMBER OF PODS AND QUALITY OF SEED HARVESTED FROM VARIOUS FLORAL UMBELS ARISING FROM VIKING BIRDSFOOT TREFOIL STEMS DURING 1955

Umbel position base to top of stem	Pods per umbel (no.)	Seeds per pod (no.)	1000-seed weight (grams)	Seed germination	
				Non-viable per cent	Live seed[2] per cent
1	2.25 [1]	13.0 [1]	1.26 [1]	11.6	80.4 [1]
2	1.32	12.8	1.20	19.8	80.2
3	0.54	11.9	1.15	26.4	73.6
4	0.17	12.5	0.82	49.8	50.2
5	0.06	12.5	0.78	56.8	43.2

[1] Duncan's multiple range test p = .05
[2] Live seed = total live seed (including hard seed)

Flowering Habit and the Timing of the Seed Harvest

In the foregoing, attention has been centred on the development of a single pod and the optimum time for harvesting it. In the following discussion consideration will be given to the related but somewhat more complicated problem of the flowering habit of the crop, its influence on the potential seed yield, and its relationship to the determination of the optimum time for seed harvest.

It is known that birdsfoot trefoil flowers profusely as the stem elongates. This flowering habit results in differential development of inflorescences, and thus in the presence of flower-buds, flowers and immature and mature pods on a stem at the same time.

In these studies differences in quantity as well as quality of seed from the various inflorescences were noted (Table 2). As many as nine flowering umbels were produced per stem. Of these nine, only the lowest five developed pods from which seed was obtained. However, of the five pod-producing umbels, only the first three produced pods from which seed of high viability was obtained. This seed was round, shiny, olive-brown to brown in colour with some seed that was slightly dented or flattened but not shiny in appearance. These characteristics, plus the fact that the seed from these lower three umbels was high in germination and seed weight, indicated that it was both physiologically and morphologically mature. Seed obtained from pods arising from the fourth and fifth umbel was green in colour and after air drying was shrunken, low in seed weight and germination percentage.

The number of umbels producing seed of high viability varied. Such variation depended upon the rate of production of successive umbels, the length of time over which the individual flowers remained open and receptive to pollinating insects, or upon a combination of these factors. The prevailing weather conditions appeared to influence this for during 1955,

a year of rapid development when temperature was high and rainfall low (Figure 1), 2 days elapsed between the development of umbels. The individual flowers remained open approximately 8 days, whereas in 1956 when temperature was lower and rainfall more plentiful than 1955, the period of time between the appearance of successive umbels was observed to be approximately 2½ to 3 days and flowers remained open and unpollinated for 9 to 10 days.

The climatic conditions which induce rapid growth and development also increase the incidence of mature pod shattering. Under these conditions not more than three umbels developed seed of similar viability before the most mature pods dehisced. These three umbels produced approximately 92 per cent of the seed that was harvested. A delay in the seed harvest to allow the fourth and fifth umbels to mature was not practical. Such a delay allowed the more mature pods of the first umbel to dehisce and resulted in a decrease in the yield by approximately 50 per cent. In addition, the number of pods per umbel which developed from the fourth and fifth inflorescences was markedly lower than those of the first three umbels even though the number of seeds per pod remained the same (Table 2).

Variations in the quantity and viability of the seed produced also resulted from the prolonged flowering time of a single plant and from the range of early to late flowering plants that occurred within a variety. The data in Table 2 consider these sources of variation. They are averages derived from harvesting pods from successive umbels of 30 randomly picked stems of Viking trefoil, which had been seeded at 2, 3, 4, 6 and 12 inches apart.

Since these data are averages over a wide range of plant diversity they are representative of the level of pod production and quantity and quality of seed as they would be found by a producer in a seed field of trefoil.

From the foregoing it is recognized that the decision as to the exact time for harvest of a seed field of this crop cannot be determined on the basis of a single developing pod nor on the indeterminate flowering habit. The decision of when to harvest must be made on the proportion of mature to immature pods.

Data from studies of the flowering habit and pod development are related to and necessary for any method which is to determine the exact time for harvest. From the studies on pod development it was found that pod colour was a convenient criterion for the determination of pod and seed maturity. Thus changes in pod colour during seed maturation could be used to estimate the maturity of the crop. The indeterminate flowering habit not only limited the maximum level of harvestable seed to that from approximately three umbels, but also necessitated that the less mature pods and flowers from the upper umbels be ignored. These data can form a basis for establishing a method for determining the proper time to begin seed harvest. Such a method involves the determination of the percentage of pods at the optimum of maturity at intervals during the development of the seed crop.

TABLE 3.—THE QUALITY OF VIKING BIRDSFOOT TREFOIL SEED HARVESTED AT DIFFERENT STAGES OF MATURITY, 1954 AND 1955 SEASONS

Most common pod colour	Per cent pods in seed maturation stage[3]		Seed maturity	Per cent of seed			1000-seed weight (grams)			Seed germination per cent: Live seed[4]		
	1954	1955		1954	1955	Av.	1954	1955	Av.	1954	1955	Av.
Green-purple	35	47	Full[2]	37.6	54.9	46.3	1.10[1]	1.44[1]	1.27	98.0[1]	96.0[1]	97.0
			Shrunken	36.8	5.4	21.1	0.69	1.08	0.89	19.2	93.0	56.1
			Shrivelled	25.5	39.7	32.6	0.38	0.56	0.47	0.0	10.1	5.0
Light green to Green-white	68	59	Full[2]	72.0	65.6	68.8	1.30	1.05	1.18	93.4	99.3	96.4
			Shrunken	22.7	4.5	13.6	0.95	0.87	0.91	34.2	94.5	64.4
			Shrivelled	5.5	29.9	17.7	0.49	0.56	0.52	9.5	19.5	14.5
Green-white to Light golden brown	76	85	Full[2]	79.3	93.4	86.4	1.37	1.25	1.31	97.4	96.0	96.7
			Shrunken	19.3	0.8	10.1	0.96	——	0.96	35.5	——	35.5
			Shrivelled	1.5	5.8	3.6	0.34	0.69	0.51	3.9	22.5	13.2
Light golden brown to Darker brown	89	94	Full[2]	76.6	91.1	83.8	1.15	1.12	1.14	97.8	96.2	97.0
			Shrunken	18.1	0.0	9.0	0.94	——	0.94	92.2	——	92.2
			Shrivelled	5.3	8.7	7.0	0.70	0.79	0.74	66.9	57.0	62.0

[1] Duncan's multiple range test p = .05
[2] Full = round, dented or slightly flattened seed
[3] Mature pods = total of green-white, light golden brown, brown and black
[4] Live seed = total live seed (including hard seed)

On this basis it was found that as the proportion of maturing seed pods increased, the amount of seed which was full or round, shiny and of high weight and germination increased (Table 3).

In order to harvest high yields of good quality seed the proportion of 60 to 85 per cent of the pods must be mature. Pod colour ranged from light green to golden brown at this time and corresponds to the time for harvest recommended by Anderson (1).

Harvesting at the 60 per cent mature pod level resulted in a relatively low return of quality seed (60-65 per cent). Since data in Figure 1 indicate that a period of 6 to 8 days occurs between the time a pod is light green in colour until it turns a golden brown, it is logical to assume that harvesting should be delayed until a higher percentage of the pods are mature. On the other hand, at the level of 85 per cent mature pods the incidence of pod dehiscence increased as the most mature pods continued to lose moisture.

It is, therefore, suggested that the harvesting of birdsfoot trefoil should be delayed a little longer than was recommended by Anderson and should begin when approximately 70-80 per cent of the pods are mature.

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PERFORMANCE OF FOUR IRRIGATED PASTURE MIXTURES UNDER GRAZING BY SHEEP[1]

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ABSTRACT

Four herbage mixtures with two levels of fertilizer application were compared for 6 consecutive years under grazing by sheep. The most satisfactory mixture with respect to yield, persistence, and stability was one of orchardgrass, *Dactylis glomerata* L., smooth bromegrass, *Bromus inermis* Leys., creeping red fescue, *Festuca rubra* L., and ladino clover, *Trifolium repens* L. The same mixture of grasses without clover was lower yielding even with annual applications of 78 pounds of nitrogen and 48 pounds of P_2O_5 per acre. A mixture of orchardgrass, tall fescue, *Festuca arundinacea* Schreb., reed canary grass, *Phalaris arundinacea* L., and alfalfa, *Medicago sativa* L. yielded well, and combated weed encroachment but the alfalfa did not persist beyond the third year. Tall fescue was very aggressive in this mixture. A mixture of orchardgrass and ladino clover yielded well for a few years but became badly infested with weeds as the orchardgrass was reduced through winter killing. Moderate yield increases were obtained from phosphorus and nitrogen fertilizer applications on all mixtures.

INTRODUCTION

Recent expansion of the irrigated acreage of southern Alberta has stimulated interest in irrigated pasture as a farm crop. This represents an elevation in status for pasture and has warranted a re-appraisal of the means of production.

The herbage mixture, being of fundamental importance, was selected as the first area of investigation. In 1946 a trial was begun to compare 24 grass-legume mixtures for yield and general suitability under a regime of clipping to simulate grazing. Four mixtures were selected from the simulated grazing trial for testing under grazing with sheep. This paper reports the dry matter yields and botanical composition of the four mixtures during the period from 1953 through 1958. Only brief reference is made to animal performance since a detailed report on that aspect will be presented in the near future.

REVIEW OF LITERATURE

Most of the current recommendations for pasture are for simple rather than complex mixtures. Simple mixtures are those containing from two to a few species but the exact definition depends on the individual and the circumstances. Hughes (3) described simple mixtures as those in which inter-species competition was not intense and used the term "ultra-simple" to refer to mixtures of one grass and one legume. The legume in such mixtures was considered a beneficial rather than a competitive associate because of its ability to introduce atmospheric nitrogen into the soil in a form usable by the grass. Mulder (6), on the other hand, expressed the view that, because of different cultural requirements, grasses and legumes could actually be competitive and suggested that in some cases fertilizers were a better source of nitrogen than were legumes.

[1]Contribution from the Forage Crops and Animal Science Sections.

Holmes and MacLusky (2) compared several strains of grasses grown in pure stands and in mixtures with clover, with and without fertilization. The grasses showed differences in degree of competition with clover, but in all cases yields of the grass-clover mixtures were higher than those of the grasses alone. It was estimated that the clover in the mixtures had a fertilizer replacement value that ranged from 28 to 205 pounds of nitrogen per acre on a dry matter yield basis.

In most methods of herbage yield determination on pasture, an attempt is made to measure the amount of herbage removed by the animal. One approach is through hand plucking or cutting of the sample area to the same degree of defoliation as that accomplished by the grazing animals (7). Another is the "difference" method in which one sample is harvested from the ungrazed and one from the grazed portion of the pasture and the difference in weight between the two samples is taken as the amount consumed by the animals (4).

MATERIALS AND METHODS

Four herbage mixtures were compared in the test (Table 1).

Annual commercial fertilizer applications were compared with a check treatment. In 1953 the commercial fertilizer treatment consisted of 100 pounds per acre of ammonium sulphate, 21-0-0, applied in July. During 1954-1956 the nitrogen treatment was changed to 100 pounds per acre of ammonium nitrate, 33.5-0-0, also applied in July. In 1957 and 1958 a further change was made to 100 pounds per acre of 33.5-0-0 applied in June and again in July. Late fall applications of 8 tons per acre of well-rotted manure were made to all plots in 1953 and in alternate years thereafter.

The experiment was laid out on soil classified as a Lethbridge loam-silty clay loam (5). Six 2-acre blocks were established, three of which were top-dressed with commercial fertilizer. Each block was then sub-divided into four half-acre plots on which four herbage mixtures were seeded at random. Each plot was further sub-divided into four eighth-acre paddocks to permit rotational grazing. The individual paddocks were 64 by 85 feet. This plan provided three replicates for fertilizers and mixtures.

TABLE 1.—MIXTURES AND RATES OF SEEDING IN POUNDS PER ACRE

Species	Herbage mixture			
	No. 1	No. 2	No. 3	No. 4
Orchardgrass, *Dactylis glomerata* L.	7	7	14	6
Smooth bromegrass, *Bromus inermis* Leys.	7	7		
Creeping red fescue, *Festuca rubra* L.	7	7		
Tall fescue (Alta), *Festuca arundinacea* Schreb.				6
Reed canary grass, *Phalaris arundinacea* L.				5
Alfalfa (Ladak), *Medicago sativa* L.				5
Ladino clover, *Trifolium repens* L.	3		3	

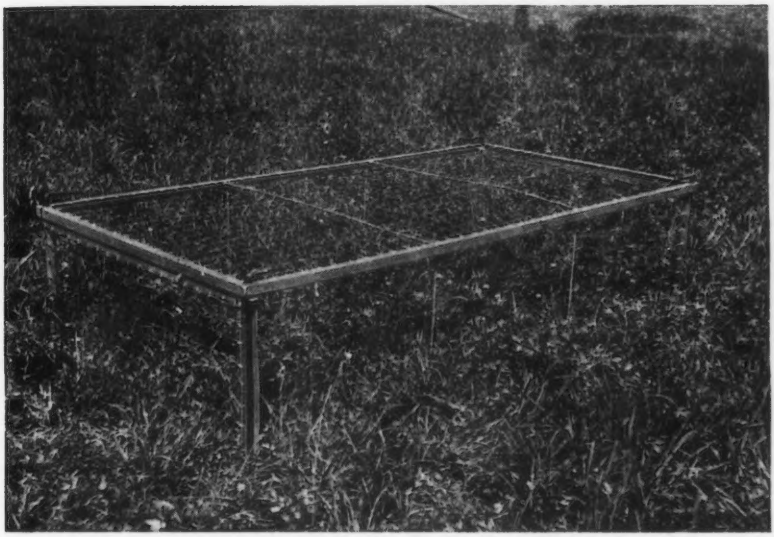

FIGURE 1. Three- by six-foot cage used to protect sample area from grazing.

Seeding was done on fallow land, without a companion crop, in April, 1952. Sprinkler irrigation was used to supplement rainfall during establishment. Satisfactory stands of the seeded mixtures were obtained on all plots after they had been mowed twice to control weeds. This growth was grazed off by dairy cattle in September.

Grazing commenced in 1953 with yearling white-faced ewes as the grazing animals. Stocking rates were established commensurate with estimated herbage yields so that all mixtures would have near equal grazing seasons. Also, an attempt was made to maintain the same animals on a plot for the entire season. It was impossible to accomplish these objectives completely and occasional adjustments in stocking rates were necessary. In spring, for example, growth began on all four rotation paddocks at the same time and a temporary surplus of herbage existed. This situation was countered by increasing stocking rates temporarily and by lengthening the grazing period. Even these adjustments were not adequate to effect complete utilization of the herbage for the sheep refused to eat the culms produced. This rejected growth was mowed following removal of the sheep and either left lying or raked up and removed, depending on the quantity. An alternative would have been to harvest the spring surplus as hay or silage for feeding later in the year, but this was impractical because of the number and size of paddocks. The average length of a rotation cycle was 1 month, with 1 week of grazing and 3 weeks of recovery for each paddock but, as already indicated, this varied with the season.

Two 3- by 6-foot protective cages (Figure 1) were used in each paddock. The four sides and top were joined by a removable pin at each

TABLE 2.—YIELDS OF THE HERBAGE CONSUMED BY SHEEP GRAZING IRRIGATED PASTURES AT LETHBRIDGE FROM 1953 TO 1958
(POUNDS OF DRY MATTER PER ACRE)

Mixture number	1953		1954		1955		1956		1957		1958		Mean 1953–1958	
	Fert.	Unfert.	Fert.	Unfert.	Fert.	Unfert.	Fert.	Unfert.	Fert.	Unfert.	Fert.	Unfert.	Fert.	Unfert.
4	5605	5447	7063	5698	6094	5132	5325	4407	7219	5374	7989	6204	6549	5227
1	4559	4292	6410	5647	6165	5370	4931	4695	7010	6104	7668	6649	6124	5460
3	4478	4519	6365	4335	5349	4595	4620	3788	6629	5414	7443	6896	5814	4924
2	4191	2614	4826	3234	4967	3400	4227	2857	6012	4165	6470	4433	5070	3450
Mean	4710	3993	6165	4729	5576	4624	4776	3937	6716	5264	7392	6046	5889	4765

The difference between fertilized and unfertilized means was significant in 1954, 1957, and 1958

	1958	1957	1954	1955	1953	1956
Fertilized	7392	6716	6165	5576	4710	4776
Unfertilized	6046	5264	4729	4624	3993	3937

Means joined by the same lines do not differ significantly at the 5 per cent level

corner, which made them collapsible for easy transport and storage. Forage yields were determined by mowing the herbage on the caged areas after a field had been grazed. The herbage was placed in paper bags, oven-dried to constant weight, and weighed. The cages were relocated at random in the next paddock of the plot and the sheep moved. The "difference" method of yield determination was followed in late spring and early summer when a considerable amount of coarse, stemmy material was left uneaten. The practice was to harvest the caged area and then cut a 3- by 6-foot strip about 6 feet away from it. The dried weight of the material harvested outside the cage was subsequently subtracted from that inside. In early spring and late summer utilization of the herbage was more complete, and sampling of the uncaged area was considered unnecessary. The practice then was to mow the herbage on the caged area to the same stubble height as the grazed portion.

Botanical composition was determined in June and September annually by hand-separation of a composite sample of green herbage weighing about 300 grams. Only one of the four paddocks of each plot was sampled but it was the same paddock each year. In addition, visual estimates of botanical composition were made at various times during the season.

In the non-clover mixture there was a constant invasion of alsike clover. This was kept under control through the use of "Brush-Kill", a mixture of equal parts of 2,4-D and 2,4,5-T in ester form applied at the rate of 20 ounces of acid equivalent per acre in 12 gallons of water. Beginning in 1955 annual treatments were made in early July. This treatment undoubtedly affected the weed population as well but it was considered necessary to preserve the difference between Mixtures No. 1 and 2. Additional weed control on all plots was accomplished by mowing whenever necessary.

Four to five flood irrigations of 2 to 3 inches each were made annually. These were supplemental to an average of 10 inches of seasonal precipitation.

RESULTS AND DISCUSSION

The average beginning and ending dates of the grazing season were May 22 and September 30, which made the average length of the season 132 days. The shortest season was 1955, with 122 days of grazing beginning on June 1. The termination of each season depended largely on the rate of stocking. Little growth occurred after the end of August so fall grazing was on growth that accumulated earlier when there was some excess over the immediate requirements.

Yields of dry matter produced each season are recorded in Table 2. Mixture No. 2, without clover, yielded the least in all years. There was no consistent difference between the other three mixtures, although in some years Mixture 3 was inferior to one or both of Mixtures No. 1 and No. 4.

Mean yields of the fertilized pastures were consistently higher than those of the unfertilized ones but the differences were significant at the 5 per cent level in only 3 of the 6 years. In all years the average yield of Mixture No. 1, unfertilized, was higher than that of Mixture No. 2,

fertilized. If the nitrogen only is considered, it can be calculated from Table 2 that the clover of Mixture No. 1 had a nitrogen fertilizer replacement value that ranged from 34 to 85 pounds per acre. This is in the lower range of the values reported by Holmes and MacLusky (2), but their swards had a higher percentage clover content.

Reasons for the differences in mean yields for the years cannot be deduced from the data presented, but consideration of the conditions prevailing each year suggest some plausible explanations. The increased yield from 1953 to 1954 would seem to be a logical progression associated with filling in of the stand and increased vigour of the plants. The decline in 1955 might be attributed in part to the spring weather conditions. Low temperatures during May permitted only slow growth and, as a result, the commencement of pasturing was delayed. This was followed by rapid June growth, which was inefficiently utilized. Yields in Table 1 do not include the herbage rejected by the sheep, which in 1955 was 21 per cent of the total yield. The 1956 yields undoubtedly were influenced by the abnormally severe winter of 1955-1956. Stands were thinned considerably and spring growth was slow. The increased yields of the fertilized pastures in 1957 and 1958 would at first suggest the influence of the change in fertilizing practice, but the accompanying increase in yield of the unfertilized pasture makes it apparent that additional factors were operating. Whatever these factors were, evidence is provided to show that pasture yields should not deteriorate in 6 years.

The amount of herbage available to the animals at any one time during the grazing season was determined by dividing the growing season into five equal intervals of 34 days beginning April 29 and ending October 15. With the beginning and ending of the growing season thus arbitrarily set, yields were calculated at 34, 68, 102, 136, and 170 days. Termination dates of the five intervals were June 1, July 5, August 8, September 11, and October 15.

Representative data are illustrated in Figure 2 and show the weight of dry matter available to the animals during each period rather than the amount produced. The difference is due to the fact that, with the four-field rotation system, though the herbage may have been produced during a certain period it was often not made available to the animals until later. Results for 1955 were selected for presentation because by that time the

TABLE 3.—PERCENTAGE OF TOTAL YEARLY DRY MATTER YIELD LEFT UNEATEN FOLLOWING EARLY SUMMER GRAZING EACH YEAR FROM 1955 TO 1958

	Mixture No. 1		Mixture No. 2		Mixture No. 3		Mixture No. 4	
	Fert.	Unfert.	Fert.	Unfert.	Fert.	Unfert.	Fert.	Unfert.
1955	13.9	17.4	21.5	25.1	18.6	20.0	22.8	25.2
1956	2.5	2.2	3.3	6.4	8.0	8.2	5.0	5.7
1957	10.0	9.0	13.0	8.2	9.7	11.6	11.9	12.0
1958	6.5	8.7	8.2	7.1	11.8	5.4	13.0	16.0
Average	8.2	9.3	11.5	11.7	12.0	11.3	13.2	14.7

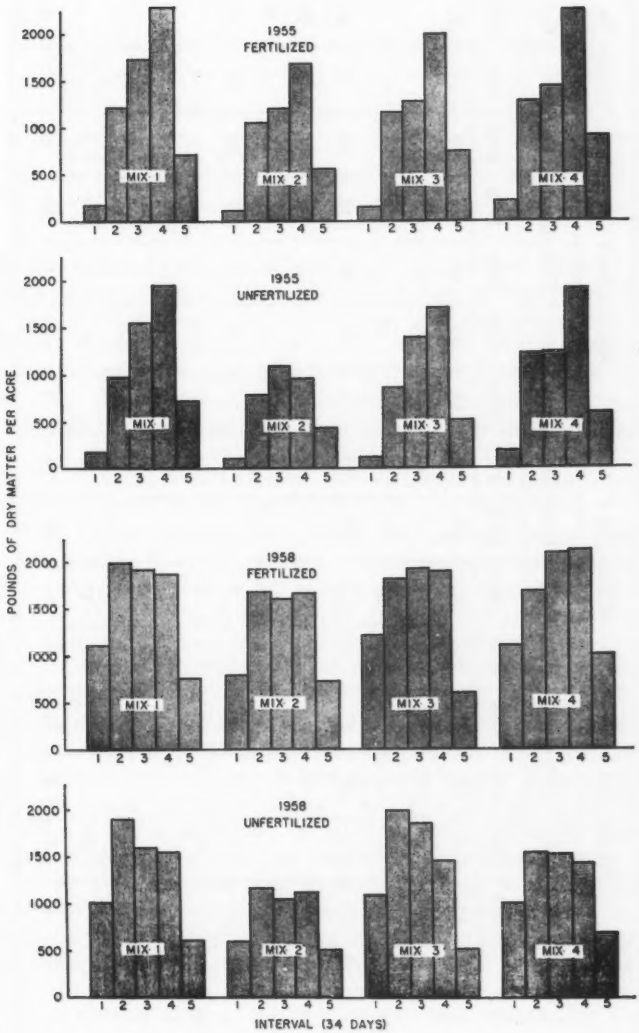

FIGURE 2. Herbage available to animals during five intervals beginning April 28 and ending October 15.

TABLE 4. — PERCENTAGE BOTANICAL COMPOSITION OF PASTURE MIXTURES IN SEPTEMBER OF 1953 AND 1958 (AVERAGE OF 3 REPLICATES)

	Mixture No. 1				Mixture No. 2				Mixture No. 3				Mixture No. 4			
	Fertilized		Unfertilized		Fertilized		Unfertilized		Fertilized		Unfertilized		Fertilized		Unfertilized	
	1953	1958	1953	1958	1953	1958	1953	1958	1953	1958	1953	1958	1953	1958	1953	1958
Orchardgrass	51	23	63	27	80	42	69	24	93	35	61	35	59	35	37	19
Bromegrass	10	20	6	12	12	7	7	12		5		5		1		1
Creeping red fescue	2	22	2	24	2	27	4	33		1		6				3
Clover	36	11	29	17	6	2	20	10	4	7	39	14	14	2	23	4
Alfalfa													12	1	20	13
Reed canary grass													5	2	9	2
Tall fescue		2		2		1		4		8		1	10	50	11	37
Kentucky bluegrass		9		5		3		2		5		9		6		5
Wild barley		T		1		14		2		14		3				1
Dandelion		10		7		4		2		14		14		3		4
Thistle		2		4		T[2]		11		11		13				11
Total weeds[1]	0	13	0	13	0	18	0	15	3	39	T	30	0	3	0	16

[1]This includes the 3 species above, plus small amounts of unlisted weeds. In 1953 the proportions of weeds were not determined by species.
[2]Trace; less than 1 per cent.

TABLE 5.—PERCENTAGES OF ORCHARDGRASS IN COMPARISON WITH OTHER GRASSES IN MIXTURES NO. 1 AND NO. 4 IN JUNE, 1955, AND JUNE, 1956

	1955	1956
Mixture 1		
Orchardgrass	32	21
Brome and fescue	47	61
Mixture 4		
Orchardgrass	29	24
Tall fescue	29	43

mixtures had attained a measure of ecological equilibrium. The 1958 results are given as a final evaluation. It can be seen that on the unfertilized plots the proportion of the total yield of herbage that was available during the latter part of the season declined from 1955 to 1958. Where fertilizer was applied, a relatively good supply of herbage was available late in the season even in 1958. This effect of fertilizer was most noticeable in the absence of a legume and could be seen on Mixture No. 2, the non-legume mixture, in 1955. The period of production varied with both the fertilizer application and the mixture, for in all years the interaction of periods x mixtures and fertilizers x mixtures was significant at the 1 per cent level.

Herbage yields given do not include the coarse, stemmy material rejected by the sheep in early summer. The amount of this material varied with the mixture and the year, but usually was a substantial part of the total yield (Table 3). The high proportion of the yield that may be in this category, up to 25 per cent, emphasizes the importance of spring management. Most of this material could be salvaged by preserving part of the early season growth as hay or silage.

Changes that occurred in species composition of the mixtures from 1953 to 1958 are presented in Table 4. Orchardgrass decreased annually in all mixtures due to winter killing. It was replaced largely by bromegrass and creeping red fescue in Mixtures No. 1 and No. 2, and by tall fescue in Mixture No. 4. The difference in survival ability of these grasses was particularly evident following the severe winter of 1955-1956. Data in Table 5 show a greater reduction in the percentage of orchardgrass than of the other grasses following that winter. When these data are considered in light of the lower yield in 1956 than 1955 (Table 2), it can be calculated that the yield of orchardgrass decreased while that of the other grasses increased. In Mixture No. 3, which included only orchardgrass and ladino clover at the time of seeding, the orchardgrass was replaced by weeds and invading grasses. Kentucky bluegrass, *Poa pratensis* L., was not listed with the weeds but it was an undesirable invader. The preference shown by sheep for orchardgrass over bluegrass was obvious. Weed populations would have been higher on all plots, but particularly on those of Mixtures No. 2 and No. 3, if it had not been for the constant effort to control them. The best defence against weeds seems to be the maintenance of a vigorous sward.

The percentages of clover were low at the time of year the determinations reported in Table 4 were made. Visual estimates, made during the season, revealed that in all years clover reached a level of about 50 per cent of the mixture in early July and then declined as the season advanced. The proportion of alfalfa in Mixture No. 4 was still as high as 65 per cent in individual paddocks in the third year, but it declined thereafter and by the fourth year its contribution to the total yield was small on most plots. Bacterial wilt, *Corynebacterium insidiosum*, was an important factor in reducing the stand. In each year the proportion of alfalfa was greatest in spring, as the sheep tended to avoid grazing it at that time in preference for the grass. They did not show this preference later in the season.

CONCLUSIONS

Assessment of the mixtures and fertilizer treatments is as follows:

Mixture No. 1

Orchardgrass, the foundation of this mixture, decreased due to winter killing but an accompanying increase in bromegrass and fescue maintained a good cover. On portions of the plots where the soil was often excessively moist the fescue first predominated, but later wild barley, *Hordeum jubatum* L., invaded. The clover reached the desirable level of 45 to 50 per cent for a brief period in late June and early July, but was too low at other times of the season. A strain of white clover that would continue to grow during late summer and a more winter-hardy orchardgrass variety would improve the performance of this mixture in southern Alberta.

Mixture No. 2

The lack of a legume made this mixture unsatisfactory. A comparison of yields in Table 2 shows that the effect due to clover in Mixture No. 1 was greater than the effect due to the fertilizer treatment on Mixture No. 2.

Mixture No. 3

This mixture provided a satisfactory yield of high quality forage in most years but it was susceptible to weed invasion following reduction of the orchardgrass stand by winter killing. This mixture contained more clover than any other mixture. It probably would be suitable for permanent pasture, if a winter hardy strain of orchardgrass was used. An improved variety of white clover would be an additional asset.

Mixture No. 4

The salient feature of this mixture was the aggressiveness of tall fescue. It gradually assumed dominance of the sward during the 6 years, particularly where fertilizer was applied. This attribute of tall fescue was desirable from the weed control aspect, but undesirable from the point of view of maintenance of other grasses and legumes. Alfalfa, for example, was almost eliminated from the fertilized plots. There was a tendency for orchardgrass to be replaced by tall fescue, probably due in part to the influence of selective grazing but, also, because of greater winter killing of orchardgrass. Reed canary grass suffered from tall fescue competition but persisted along

ditch banks and in low wet spots where competition was less severe. Yields of this mixture were relatively high in most years and it was not difficult to manage. On the basis of observations throughout the experiment, it was concluded that this mixture would be better adapted to excessive soil-moisture conditions than would any of the others tested.

Fertilizer Treatments

Although yield increases from fertilizers were not great, their potential to increase yields was indicated. Also, the fertilized pastures required less mowing to control weeds, a factor of some economic importance. The role of fertilizers is now under more critical investigation.

General

Under good management irrigated pastures can remain productive for several years. Yields of all mixtures were higher in the sixth year than in any previous year. The fact that there was little difference in the yields of the three better mixtures is verification of the results of the original clipping test.

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HOST RANGE STUDIES OF THE LITTLE CHERRY DISEASE VIRUS[1]

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SUMMARY

Attempts to pass the virus of the little cherry disease through apricot and Italian prune have been unsuccessful. Some clones of the native cherry, *Prunus emarginata* var. *mollis* have been shown to carry little cherry virus without symptoms, but other clones have been demonstrated immune. *P. emarginata* var. *mollis* is a possible source for the original introduction of little cherry virus into sweet cherry.

INTRODUCTION

Recent reports of the presence in oriental flowering cherries of a virus that induces little cherry disease symptoms in sweet cherry (4, 5) have renewed interest in the host range of the virus causing the little cherry disease in the Kootenay region of British Columbia.

Natural occurrence of the disease has been reported only from this region. The course of its rapid spread was followed by extension personnel from 1933, when it was found in one orchard at Willow Point, British Columbia, until 1948 when all cherry trees in most Kootenay cherry-growing districts were diseased. Such rapid progress of a disease from one known source orchard stimulates speculation on the means by which the virus was introduced to sweet cherry in this particular location.

The disease has been reported by Foster and Lott (1) to cause symptoms in sweet cherry (*Prunus avium* L.) and sour cherry (*P. cerasus* L.). Transmission was not demonstrated in transfers of buds from shrubs of wild cherry (*P. emarginata* L. var. *mollis* (Dougl.) Brewer) growing near affected orchards. Wilks and Milbrath (6) have shown that the causal virus does not infect peach (*P. persica* Sieb. & Succ.) and western choke-cherry (*P. virginiana* L. var. *demissa* (Torr. & Grey) Torr.), although these are alternate hosts of the western X-disease virus, the cause of small cherry symptoms in sweet cherry in other regions (2, 3). Wilks and Reeves (7) have reported the occurrence of a virus that is latent in flowering cherry, and that, by means available, cannot be distinguished from the little cherry virus. Also, they reported that flowering cherry trees that were growing in 1933 in the district where the little cherry disease originated are still living and are now infected with little cherry virus.

Additional host range studies of the little cherry disease, thus far unreported, were conducted in the years 1946-1950, but limited availability of healthy bearing test cherry trees restricted their scope. Demonstration that the cherry varieties Sam and Star are foliage indicators for little cherry virus (8) made possible, in the years 1958-60, amplification of the earlier tests. The present paper reports the results of transmission tests which were designed to determine whether apricot, Italian prune and *P. emarginata* var. *mollis* can serve as alternate hosts.

[1]Contribution No. 17 from Research Station, Canada Department of Agriculture, Summerland, B.C.

EXPERIMENTAL PROCEDURES AND RESULTS

Apricot

Buds from affected sweet cherry trees were applied to one bearing tree each of Wenatchee Moorpak and Blenheim apricot in August, 1946, using 10 inoculum buds per test tree. These trees were re-budded in 1947 and 1948. In each season, several of the buds remained alive until the following spring. The inoculated trees displayed no symptoms that could be attributed to the little cherry virus.

In the spring of 1959, 8 of a group of 15 apricot seedlings were inoculated with little cherry virus, using 3 buds per tree. No symptoms developed in the inoculated seedlings in 1959 or 1960. In the spring of 1960 these inoculated trees were indexed on the indicator variety Sam. No foliage symptoms developed during the summer or fall, indicating that none of the apricot seedlings carried the little cherry virus.

Italian Prune

Two Italian prune trees received 10 buds each from infected sweet cherry trees in 1946, 1947, and 1948. Prune dwarf symptoms appeared on inoculated branches of both trees in 1947, but no symptoms attributable to little cherry virus resulted from these tests. Two bearing sweet cherry trees received 8 buds each from the inoculated Italian prune trees in 1948 and 1949. Little cherry symptoms did not appear in these test trees.

In the spring of 1959, three young prune trees received 3 buds each from an infected sweet cherry source. Two trees of the same age served as uninoculated checks. No symptoms developed in the test trees in 1959 or 1960. The inoculated prune trees were indexed on test trees of Sam cherry. No foliage symptoms had appeared by the end of the 1960 growing season, indicating that the prune trees had not acquired little cherry virus.

Prunus emarginata var. *mollis*

Buds from infected sweet cherry trees were applied to six bushes of this wild cherry species in August, 1946. The procedure was repeated in 1947 and 1948. No disease symptoms resulted although the sweet cherry buds made union and grew. Indexing of these test shrubs on bearing sweet cherry test trees in 1948 and 1949 yielded negative results.

A second attempt was made to inoculate this species in 1951, using four bushes that had been transplanted into a test plot. They received buds from diseased sweet cherry in August, 1951 and again in August, 1952. Although bud union was obtained in both years no symptoms attributable to little cherry virus appeared on the test shrubs. Subsequent indexing of these inoculated shrubs on Star cherry in 1953 indicated that the little cherry virus was not present in them.

P. emarginata var. *mollis* growing in or near diseased cherry orchards has been indexed for presence of the virus. In both 1946 and 1947, three healthy bearing Lambert cherry trees received buds from a group of *P. emarginata* var. *mollis* bushes growing at the edge of a Willow Point sweet cherry orchard that had been infected for 11 years. In 1947, two of the

three test trees bore fruits with little cherry symptoms, on the inoculated branches only. In 1948 all fruits on these trees were affected. The third inoculated tree and four uninoculated Lambert trees in the same planting bore normal fruits in both seasons.

In August, 1949, budwood from four sources of *P. emarginata* var. *mollis* was applied to healthy bearing Lambert cherry trees growing under screen cages. The sources included one of the bushes at Willow Point for which the earlier indexing had been positive, bushes growing at the edges of diseased orchards in two other Kootenay districts, and a branch of wild cherry growing on a diseased Lambert cherry tree in an experimental orchard. This branch had grown from a bud applied by T. B. Lott in the course of little cherry transmission tests in 1942. The caged tree that had received buds from the wild cherry source used in the 1946-47 tests produced fruits with little cherry symptoms in 1950, thus confirming the earlier transmissions from this source. The caged test trees that had received buds from the three other wild cherry sources failed to produce little cherry symptoms in 1950 or ensuing years, although the wild cherry buds made good union, and although buds from the same sources were applied in 1950.

In August, 1955, seven Star cherry trees in a screen cage received buds from *P. emarginata* var. *mollis* bushes growing in the same location as those from which transmission had been obtained in earlier tests. Each test tree received buds from a single source shrub. Five of these test trees showed foliage reddening in 1956 and thus provided additional evidence for presence of little cherry virus in shrubs at this location. The other two test trees did not show symptoms in 1956 or in subsequent years.

In the spring and summer of 1959, the sample indexing of *P. emarginata* var. *mollis* bushes was expanded, with collection of budwood from 11 bushes of this species, each from a different location, but all growing in the vicinity of infected sweet cherry orchards in West Kootenay districts. Each source bush was indexed on two trees of Sam cherry established in a screenhouse. The test trees that were used for indexing of 6 of the 11 bushes have shown foliage symptoms, indicating that these *P. emarginata* var. *mollis* sources are carrying little cherry virus.

The results of host range studies of *P. emarginata* var. *mollis* are summarized in Tables 1 and 2. Eleven clones that have been inoculated with little cherry virus have failed to become infected (Table 1). Nine bushes of this species growing near diseased cherry trees have indexed free from the virus (Table 2). Nevertheless, the presence of little cherry virus in bushes growing in the one location at Willow Point has been demonstrated in three successive tests (Table 2). Moreover, presence of little cherry virus has been demonstrated in bushes from six other locations.

DISCUSSION

The experimental evidence that little cherry virus does not produce symptoms in apricot or prune is supported by observations in commercial plantings throughout the Kootenay region, where these species are inter-

TABLE 1.—ATTEMPTED INOCULATION OF WILD CHERRY (*P. emarginata* VAR. *mollis*) WITH LITTLE CHERRY VIRUS

Year inoculated	Year indexed	Number inoculated	Number from which virus recovered
1942[1]	1949–50	1	0
1946–48	1948–49	6	0
1951–52	1953	4	0

[1]By T.B. Lott in earlier investigations (1)

TABLE 2.—INDEXING OF WILD CHERRY (*P. emarginata* VAR. *mollis*) BUSHES GROWING NEAR INFECTED ORCHARDS

Year indexed	Number indexed	Number positive
1946–47	1	1*
1949–50	3	1*
1955	7	5*
1959	11	6

*From single location at Willow Point

planted with diseased sweet cherry. The experimental evidence indicates that both apricot and prune are immune rather than tolerant.

Negative results from attempts to inoculate 11 clones of the wild cherry *P. emarginata* var. *mollis* indicate that at least some clones of this species are immune to the virus that causes the little cherry disease in sweet cherry. The apparently contradictory demonstrations, that wild cherry bushes growing in seven Kootenay locations carry a virus that causes little cherry symptoms in sweet cherry, have two possible explanations.

The most probable explanation is that, although some clones of the species can become infected with little cherry virus, other clones are immune. As all bushes are seedlings, and as there are obvious morphological variations within the species and its varieties, heterogeneity for virus immunity is quite conceivable.

These results could also be explained by the existence of two distinct viruses capable of inducing little cherry symptoms in sweet cherry, the one occurring in some bushes of wild cherry, the other widespread in sweet cherry but unable to infect wild cherry.

Knowledge that *P. emarginata* var. *mollis* can carry little cherry virus renders less promising any program for eradication of the disease in the Kootenays by removal of infected sweet cherry plantings. On the other hand, the addition of this species to the little cherry virus host range will have no serious influence on control programs that may become necessary in other major cherry-growing districts of British Columbia, because the distribution of this native cherry reaches these other districts only sparsely, at several scattered points.

In speculation concerning introduction of the little cherry disease to Kootenay cherry plantings, *P. emarginata* var. *mollis* must be considered

as a possible source of the virus. It grows abundantly in the district first invaded by the disease. The bushes from which the virus has been transmitted repeatedly are growing at the edge of one of the first three sweet cherry orchards in which little cherry disease was recognized. Thus, at the time sweet cherry was first affected at Willow Point, two symptomless hosts, wild cherry and flowering cherry, were growing near by. Either or both may since have been infected by natural spread. Alternatively, either could have been the source of the virus that was introduced into sweet cherry.

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ROW WIDTH AND SEEDING RATE IN RELATION TO SEED PRODUCTION IN TIMOTHY, *Phleum pratense* L.[1]

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ABSTRACT

Row widths of 14, 21, 28 and 35 inches gave similar seed yields, outyielding the 7-inch row width by 109 pounds per acre over a 4-year period. Row width had only minor effects on seed quality measured by seed weight, percentage of seed that established normal seedlings in soil and early seedling height. Rows 21 inches and wider required weed control measures. Based on seed yield, seed quality and weed control, the 14-inch row spacing was superior.

Seeding rates of 2.5, 5, 7.5 and 10 pounds per acre had no important effects on seed yield or seed quality. Of the three yield components studied — spike number, spike length and seed weight — only the last two were correlated with seed yield. Spike number was not related to seed yield in any of the 3 years in which it was studied but may have been important in determining differences among years in seed yield. None of the indices of seed quality was related closely to seed yield.

INTRODUCTION

The increased use of pedigreed seed of timothy in Ontario has focused attention on the need for information on seed production techniques in this crop. Ontario has been the major producer of timothy seed in Canada through the years. Basically this production has been obtained from surplus hay acreage and not from fields seeded specifically for seed production.

Stabilization in the volume of seed produced annually and in seed quality is required for the successful incorporation of varieties into a crop production program. This can be attained only when the basic volume of seed is produced from stands seeded specifically for seed production, rather than as a by-product of hay production. This means that, if the timothy seed crop is to be grown by seed growers in Eastern Canada, it must compare favourably in potential returns per acre with other competing feed and cash crops. Information on methodology in seed production is needed to place this crop in as favourable a competitive position as possible. This investigation was designed to study row width and rate of seeding as factors influencing seed yield and quality.

Studies reported on row width and rate of seeding summarized by Fulkerson (2) indicate that little information is available on the effect of these factors on grass seed production in the humid regions of Eastern North America. Buller *et al.* (1) in Pennsylvania reported Climax timothy seed yields from broadcast seedings at 6 and 3 pounds per acre and from row plantings at 3 and 1½ pounds per acre. Row plantings and light seeding rates were superior. Rate of seeding was less important than row planting in determining seed yield. Several studies in Scandinavia and Germany were summarized by Schwanbom and Froier (3). Row plantings were superior to broadcast seedings, except in Finland where both were equal. Narrow rows, 14 to 20 inches, were better than wide rows.

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TABLE 1. — TIMOTHY SEED YIELD, SEED QUALITY AND YIELD COMPONENT DATA FROM FIVE ROW WIDTHS EACH AVERAGED OVER FOUR RATES OF SEEDING AT GUELPH

Year	Row width in inches						L.S.D. 0.05	C.V.
	7	14	21	28	35	Mean		
	Seed yield in pounds per acre							
1955	278	352	383	358	336	341	58	9.1
1956	244	361	336	357	320	324	54	9.4
1957	189	307	296	413	370	315	46	11.5
1958	76	125	196	201	194	158	75	12.7
Mean	197	286	302	332	305	285	50	4.9
	Number of spikes per square foot							
1956	54.4	64.9	48.2	44.8	36.8	49.8	10.7	9.7
1957	40.9	69.2	47.8	46.3	43.2	49.6	8.6	15.4
1958	24.2	23.2	27.3	28.3	22.6	25.1	N.S.	14.1
Mean	39.9	54.2	41.1	39.7	34.0	41.8	3.1	8.0
	Spike length in centimetres							
1956	6.5	6.8	6.8	7.1	7.1	6.9	0.3	4.3
1957	6.7	7.3	7.7	8.8	8.6	7.8	0.9	6.7
1958	6.9	6.8	7.2	7.1	7.5	7.1	N.S.	6.1
Mean	6.7	7.0	7.2	7.7	7.7	7.3	0.3	4.5
	1000-seed weight in milligrams							
1955	304	318	331	352	343	330	16	5.4
1956	346	351	371	367	364	365	17	4.9
1957	346	345	359	371	370	358	9	6.2
1958	357	346	352	352	369	355	20	3.1
Mean	338	340	353	361	368	352	2	2.8
	Per cent establishment							
1955	65.9	63.3	63.8	55.1	61.1	61.8	N.S.	9.7
1956	73.0	66.0	72.5	72.5	68.7	70.5	N.S.	7.3
1957	78.2	72.0	71.5	74.5	70.2	73.3	3.3	9.5
1958	79.8	77.0	78.8	78.7	80.2	78.9	N.S.	4.7
Mean	74.1	69.6	71.6	70.2	70.1	71.1	N.S.	3.0
	Seedling height at 45 days in centimetres							
1955	17.9	17.6	16.1	18.6	16.5	17.3	N.S.	14.4
1956	10.0	9.7	9.8	10.2	10.0	9.9	N.S.	9.7
1957	9.7	9.7	8.5	8.0	8.1	8.8	0.6	6.7
1958	8.3	7.3	8.0	7.8	8.0	7.9	N.S.	4.0
Mean	11.5	11.1	10.6	11.1	10.7	11.0	N.S.	6.0

MATERIALS AND METHODS

Medon timothy was seeded in August 1954 at four seeding rates, i.e. 2.5, 5, 7.5 and 10 pounds per acre in five row widths, i.e. 7, 14, 21, 28 and 35 inches. Plot size, experimental design and plot management were the same as reported previously in a similar study with orchardgrass (2). The only difference was that three replications were used in this timothy study.

Seed yields are reported on the basis of pounds of cleaned seed per acre for the 4-year period of 1955-1958. Measurements of three components of yield were taken. Fertile spikes per square foot were estimated from counts of fertile spikes per foot of row, using four samples per plot. Fifty spikes per plot were measured to determine spike length and 200 seeds per plot weighed to estimate weight per 1000 seeds. Spike number and spike length measurements were taken in 3 of the 4 years, 1956-1958.

Seed weight, percentage establishment and seedling vigour were used to assess treatment effects on seed quality in each of the 4 years. The latter two measurements were made from a greenhouse seeding where 100 seeds per treatment were seeded in a medium-textured soil at a depth of ½-inch in a 4-inch clay pot. Per cent establishment represents the percentage of seed that established normal seedlings, and seedling height at 45 days was used as an index of seedling vigour.

EXPERIMENTAL RESULTS

Seeding rate had little effect on the attributes of seed production studied. The only data involving seeding rate reported are for those cases where the seeding rates responded differently.

Seed Yields

Row width had pronounced effects on seed yield (Table 1). The greatest difference occurred between the 7-inch row spacing and the wider rows. This amounted to 109 pounds per acre in favour of the 7-inch rows over the 4-year period. Seed yields showed both linear and quadratic responses to variations in row width (Table 2) with yields increasing as row width increased. The 7- and 35-inch rows produced yields lower than expected for a completely linear response curve. The row width effect was not the same in all years. In the first 2 years no yield differences were found among the 14- through 35-inch rows. But in the third year,

TABLE 2. — "F" VALUES FOR THE LINEAR AND QUADRATIC COMPONENTS OF THE VARIANCE FOR ROW WIDTH EFFECTS ON TIMOTHY SEED YIELD, COMPONENTS OF YIELD AND SEED QUALITY. THE DATA ARE AVERAGED OVER FIVE RATES OF SEEDING AND 4 YEARS

	Degrees of freedom	Seed yield	Spike numbers[1]	Spike length[1]	Weight of 1000 seeds	Per cent establishment	Seedling height
Row widths	4	14.2**	11.2**	8.5**	17.1**	3.1	1.5
Linear	1	31.1**	14.6**	30.4**	66.5**	7.5*	2.6
Quadratic	1	20.2**	8.9*	1.4	0.1	1.0	0.2
Deviation from reg.	2	2.7	10.6**	14.4**	30.9**	2.0	1.5

[1]Mean of 1956-1958 data

the 14- and 21-inch rows were lower yielding than the 28- and 35-inch rows, and in the fourth year the 14-inch row produced lower yields than the three wider row spacings.

Seeding rate affected yield only in the first crop year. In that year seed yields were 364, 337, 344 and 321 pounds per acre at the 2.5- through 10-pound rates, respectively. The lowest rate was superior to the two intermediate rates, which were similar in response. The 10-pound rate produced the lowest seed yield. No row width x seeding rate interaction involving yield was obtained.

Fertile Spikes

The numbers of spikes per square foot producing seed (Table 1) were similar in 1956 and 1957. The 14-inch rows produced the most fertile spikes. As row spacing increased from 14 to 21 inches a sharp decline in spike number occurred. Further increase in row width caused little if any decline in spike number. In 1958, the fourth year of production, no differences were found among row widths. Over the 3 years (Table 2) the response was not completely linear, the deviation from linearity being due to the relatively low number in the 7-inch rows and high number in the 14-inch rows. Seeding rate did not influence the number of seed producing spikes.

Spike Length

Spike length increased in a linear manner (Table 2) as row width increased. The response was greatest in 1957 as shown in Table 1. In this year spike length increased markedly and relatively uniformly as row spacing widened to 28 inches. The 28- and 35-inch row widths gave similar spike lengths.

Seeding rate influenced spike length in 1957 where a row width x seeding rate interaction occurred. This interaction, illustrated in Figure 1, indicates that low seeding rates compared with high rates performed best in narrow rows. For example, in the 14-inch rows the 2.5-pound rate was superior to the 7.5- and 10-pound rates, while in the 28-inch rows it was inferior to the 7.5-pound rate and equal to the 10-pound rate. Similarly, the 5-pound rate, which was equal to the higher rates in 14-inch rows, was inferior to both higher rates in the 28-inch rows. The difference between the 2.5- and 5-pound rates was greater at the 14-inch rows than at the 28-inch rows. Further, the two high rates, in contrast to the low rates, showed a reduction in spike length as row width increased from 28 to 35 inches.

Seed Weight

Seed weights were similar in 1956, 1957 and 1958 and slightly above the seed weight of the first crop year (Table 1). Over the 4-year period seed weight showed a linear response to row width (Table 2) with the heaviest seed being produced on the wide rows. However, it is apparent that year effects occurred. Row width had the most pronounced effect in the first crop year where seed weight was higher at each successive row width up to 28 inches. It had the least effect in the fourth crop year.

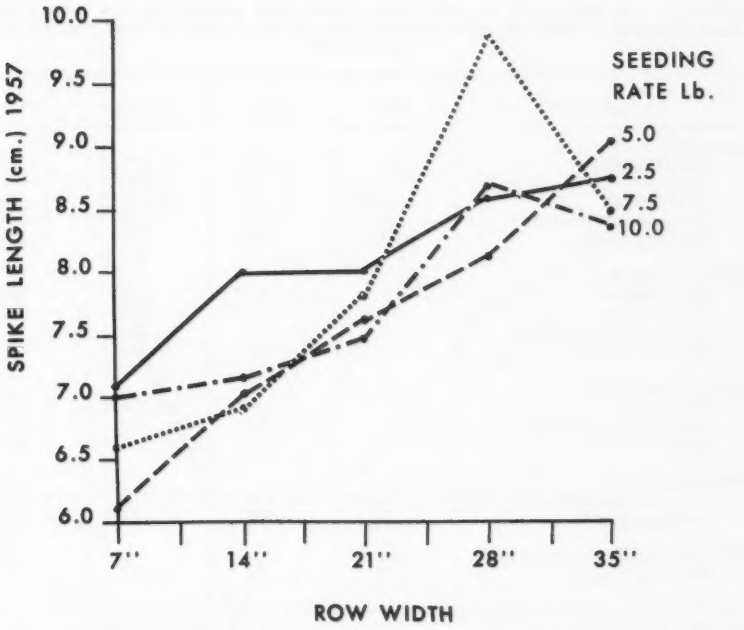

FIGURE 1. Spike length in 1957 obtained from timothy seeded in 1954 at four rates in five row widths at Guelph.

Establishment and Vigour

The two cultural practice variables had relatively unimportant effects on seed quality as measured by the percentage of seed that established normal seedlings and by the height of the seedlings 45 days from seeding. Percentage establishment declined as row spacing increased (Table 2) but the absolute amount was small (Table 1). Seedling height differences occurred in 1957 and in 1958, but the extent of these differences was small. In 1957, the 7- and 14-inch rows were superior to the wide rows. In 1958 a row spacing x seeding rate interaction was found. This interaction, illustrated in Figure 2, indicates that, although all rates produced equally vigorous seedlings at the narrow row spacings, the two lowest rates were superior in the wide row spacings.

Interrelationship of Characters

Of the three yield components – spike number, spike length and seed weight – studied in 3 years, only the last two were correlated with seed yield (Table 3). These relationships were positive. Spike length showed the closest relationship with seed yield in that the correlation coefficients were significant each year and higher than those involving seed weight in each year. Only one of the correlation coefficients for seed yield with

TABLE 3. — CORRELATION OF SEED YIELD, SEED YIELD COMPONENT AND SEED QUALITY MEASUREMENTS IN TIMOTHY GROWN IN FIVE ROW SPACINGS EACH AT FOUR SEEDING RATES AT GUELPH

Correlation of:[1]	1955	1956	1957	1958
Seed yield with:				
Spike number	—	+.029	+.075	+.389
Spike length	—	+.536*	+.804**	+.601**
Seed weight	+.438*	+.240	+.654**	+.115
Per cent establishment	−.243	−.216	−.358	−.068
Seedling height	+.009	−.064	−.716**	−.044
Spike number with:				
Spike length	—	−.444*	−.194	−.062
Seed weight	—	−.757**	−.385	−.365
Per cent establishment	—	−.217	−.240	−.438
Seedling height	—	−.261	+.396	+.245
Spike length with:				
Seed weight	—	+.468*	+.681**	+.498*
Per cent establishment	—	−.041	−.208	+.373
Seedling height	—	−.249	−.735**	−.038
Seed weight with:				
Per cent establishment	−.547*	+.025	−.025	+.522*
Seedling height	+.098	−.037	−.724**	+.139
Per cent establishment with:				
Seedling height	−.383	+.361	+.442	−.015

[1]n = 18
*r exceeds p = 0.05 (.444) and **r exceeds p = 0.01 (.561)

spike length and with seed weight was a high value. This was a value of +.804 for the correlation of seed yield with spike length in 1957. Although no relationship was detected between spike number and seed yield, spike number appeared to be negatively associated with the two other yield components, spike length and seed weight in one year, 1956. Spike length and seed weight were positively associated in each of the 3 years studied.

None of the indices used to describe seed quality — seed weight, percentage establishment and seedling height — was closely related to seed yield. Seed weight showed some relationship in that all correlation coefficients were positive and two were significant. Seedling height was negatively associated with seed yield in one of the 4 years, and percentage establishment showed no relationship to yield. Spike number, one of the components of yield, was correlated negatively with seed weight in 1956 and in the other 2 years the values were negative but below the level of significance. It was not related to percentage establishment or seedling height. Spike length, another yield component, was related positively to seed weight in each year, not related to percentage establishment, and negatively associated with seedling height in only one year, 1957. Seed weight, a component of yield and as well one index of seed quality, was variable in its relationship to the other two indices of seed quality. It was negatively associated with percentage establishment in 1955 and positively associated

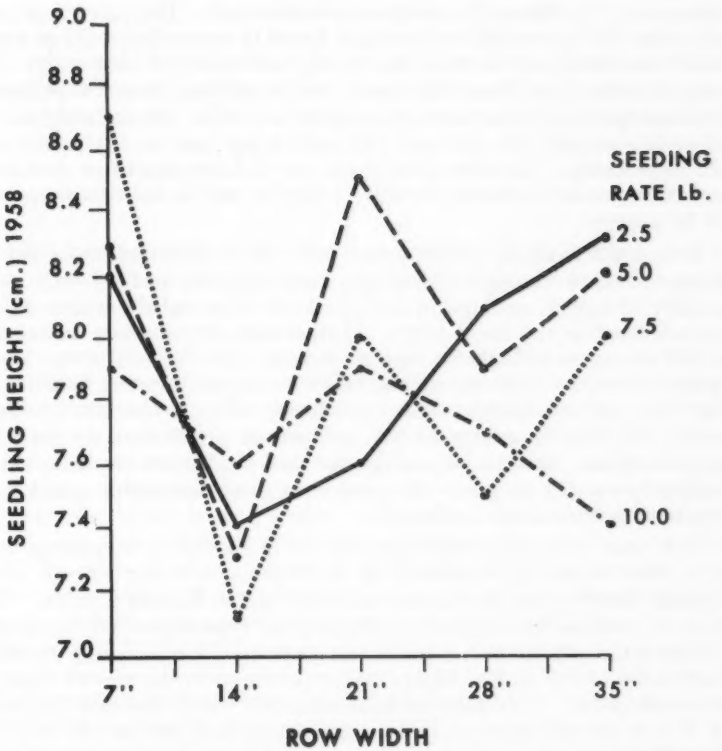

FIGURE 2. Seedling height (45 days) from seed produced in 1958 from timothy seeded in 1954 at four rates in five row widths at Guelph.

in 1958. Similarly, it was correlated to seedling height in only one of the 4 years, 1957, and the relationship was negative. Percentage establishment and seedling height correlation coefficients were not significant although the value approached the 0.05 level of significance in 1957 and was positive.

DISCUSSION

In each of the first 3 crop years the seed yield levels were similar and were moderately high. The yield level dropped by approximately 50 per cent in the fourth crop year. It is not possible to determine the separate effects of age of stand and season on seed yield from this study. However, 1958 was a dry season with the crop showing evidence of severe moisture stress in June. The accumulated April-June precipitation was 3.67 inches in 1958 in contrast to 10.94 inches in 1957, 10.42 inches in 1956 and 8.81 inches in 1955. It is likely that most of the sharp decline in average yield in 1958 was due to environmental conditions rather than age of stand. No

evidence of a "sod-bound" condition was observed. The pattern of seed yield in the first 3 years differs from that found in orchardgrass (2) in a trial seeded immediately adjacent to the timothy and managed identically. Although the data from these trials could not be analysed as one experiment, general comparisons between the two species are valid. In orchardgrass the seed yield averaged 119, 270 and 217 pounds per acre in 1955, 1956 and 1957, respectively. The low yield from the orchardgrass in its first crop year is in contrast to timothy which yielded as well in the first crop year as in later years.

Row width is clearly an important factor in determining seed yields in timothy. The 14- through 35-inch rows were superior to the 7-inch rows, essentially broadcast seedings, in each of the 4 years and the widest differences occurred in the last 2 years. Furthermore, the absolute amount of the difference was sufficiently high to indicate that the seed crop should be grown in rows. This fact means that best seed yields cannot be obtained under the current system used in Ontario where broadcast seedings, primarily the surplus acreage of hay and pasture production, are used for seed production. Specialized seedings for seed production are indicated as in orchardgrass (2) to place this crop in a more favourable position to compete with other crops in Ontario.

The extra yield, 109 pounds per acre, from all wide row spacings over 7-inch rows, could be important in determining whether or not some pedigreed timothy seed production will continue in Eastern Canada. The volume of certified timothy of the Climax plant type is presently produced in Western Canada and this is likely to continue. However, it is desirable to have some of the seed of higher classes produced in the area of timothy seed consumption. This is desirable, at least until studies indicate that plant type is not altered appreciably by seed production out of the area of consumption.

The data presented show that no differences in seed yield existed among the 14- through 35-inch rows on the average over the 4-year period. One important observation recorded was that no weed control measures were required for the 14-inch row treatment but that weed control was required in the wider rows. In the light of this, the 14-inch row has a production advantage. On the other hand, it is significant also that, although 14-inch rows were as high yielding as wider rows on the 4-year average and in the first 2 years, the 28-inch row outyielded the 14-inch row by 91 pounds per acre in the last 2 years. This suggests that wider rows might be useful for long-term stands in situations where low-cost weed control practices are available. An advantage for wide rows was reported also in orchardgrass (2), although the row width x year interaction effects were greater in orchardgrass. However, no conclusion can be drawn on a possible advantage of wide rows for long-term stands because effects of length of stand and season cannot be separated in this study.

Row width changes affected the components of yield differentially. As row width increased yield increased, although not in a completely linear manner. The two yield components – spike length and seed weight –

increased in a linear manner and were correlated positively, although not highly correlated except in 1957, with yield. Spike number increased as row spacing moved from 7 to 14 inches, then declined with wider row spacing. However, no significant correlation coefficients were found between spike number and seed yield. At a certain low level of spike numbers it is obvious that spike number is an important factor determining yield. It would appear that in this study the treatments did not depress or increase spike number far enough for it to be a major differentiating component of yield.

Years had a major effect on spike number. In 1958, a season low in rainfall from April through June, the level of spike number dropped to approximately 50 per cent of the previous level in 1957 and 1956. The other two yield components – spike length and seed weight – did not decline, yet seed yield declined by approximately 50 per cent. It would seem likely that this degree of difference in spike number was reflected in yield. Differences approaching 50 per cent in spike numbers occurred in only two cases within a year. The 14-inch row compared to the 35-inch row in 1956 was 43.3 per cent higher in spike number and had 11.4 per cent higher yield even though it was 4.2 per cent lower in spike length and 3.5 per cent lower in seed weight. The other case involved a difference of 40.9 per cent in spike number and occurred between the 7- and 14-inch rows in 1957. It would appear that the threshold in spike number per square foot, below which spike number became a major determining component of yield, was somewhere between 25 and 40 spikes per square foot. This study, of course, cannot fix the threshold precisely. It is significant to note that the relationship between yield and number of seed producing inflorescences and between yield and seed weight were not the same in timothy as reported in orchardgrass (2).

Row spacing exerted its greatest effect on seed yield and had much less important effects on seed quality. In general, seed weight and percentage of seed that established normal seedlings increased slightly as row width increased. No effect on seedling vigour was detected except in one year and here the 7- and 14-inch row widths produced the most vigorous seedlings. The 14-inch row width produced seed satisfactory in the three attributes of seed quality.

Environment was good for establishment when the trial was seeded and excellent stands were obtained, even at the lowest seeding rate. At the resulting levels of plant population seeding rate had little effect on seed yield. This finding is similar to that reported by Buller *et al.* (1) in timothy where a light rate was superior, but only slightly so, to a moderate rate. The only effect of seeding rate observed in the present study occurred in the first crop year where the lower rates were superior. This is contrary to that found in orchardgrass (2) where although, as in timothy, seeding rate affected yield only in the first crop year, heavy rates were superior. Seeding rate had no important effect on seed quality. The only other effects of seeding rate involved a row width x seeding rate interaction in spike length in 1957 and in seedling height in 1958.

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HONEYBEE ACTIVITY ON SAFFLOWER
(*Carthamus tinctorius* L.)

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ABSTRACT

Honeybees (*Apis mellifera* L.) visited safflower (*Carthamus tinctorius* L.) for nectar and pollen. About 90 per cent of the honeybees observed visited the crop in the morning. The nectar contained about 25 per cent sugars. Honeybees were by far the predominant visitors although other bees also visited the flowers. Safflower plots to which insects had full access produced twice as much seed as plots from which insects were excluded during the blooming period.

INTRODUCTION

Safflower (*Carthamus tinctorius* L.), an edible oil seed crop, has been grown since ancient times in India and the Middle East. Because of the increasing market for industrial and edible oils, this crop is now grown commercially in North America (3, 5). For this reason it was deemed desirable to investigate the value of this species as a food source for honeybees. Moreover, since safflower is a partially cross-pollinated crop (2), some information could be obtained on the effect of insects on pollination and seed production. Following is an account of some preliminary observations.

MATERIALS AND METHODS

This work was carried out in 1958 on a 1-acre planting of "Selection Ottawa 8" safflower. This planting on the Central Experimental Farm, Ottawa, was under study by A. G. Plessers, Genetics and Plant Breeding Research Institute, Canada Department of Agriculture.

Shortly before the safflower began to bloom, pairs of adjacent plots were staked out on three locations of the experimental field, and one plot in each pair was caged with transparent plastic screening to exclude insects. After the blossoming period the cages were removed. The six plots were harvested separately by hand, and the amount of clean seed weighed.

To estimate the insect abundance on safflower, five 4-square-yard plots were selected at random. On August 4-7 and 12, when safflower was at the peak of bloom, insect visitors were counted on the hour from 6.00 a.m. to 6.00 p.m., E.S.T. It was recorded how many of the honeybees and bumblebees were collecting pollen. A 1-minute observation was made on each counting station.

To estimate the nectar sugar concentration of safflower, honeybees were captured as they were foraging nectar on portions of the field where neither counting stations nor seed plots were located. These honeybees were dissected and the per cent total solids of the nectar in the honey-stomachs read on a portable Zeiss-Abbé refractometer.

RESULTS AND DISCUSSION

The observations made during the peak of the blooming period of safflower indicated that honeybees were by far the most numerous visitors

TABLE 1.—TOTAL NUMBER OF INSECT VISITORS OBSERVED HOURLY ON FIVE 4-SQUARE-YARD PLOTS (AUGUST 4-7 AND 12, 1958) AS RELATED TO NECTAR SUGAR CONCENTRATION

Hour E.S.T.	Honeybees	Native bees	Other insects	Average nectar sugar concentration
6– 7 a.m.	1	6	0	12.7% (n= 7)
7– 8	63	6	1	16.8% (n= 62)
8– 9	254	10	3	26.1% (n=129)
9–10	425	9	4	29.5% (n=136)
10–11	432	5	6	28.6% (n=129)
11–12	255	4	9	25.9% (n= 96)
12– 1 p.m.	99	2	15	23.8% (n=107)
1– 2	39	2	16	23.8% (n= 54)
2– 3	7	1	15	24.0% (n= 24)
3– 4	6	1	23	24.7% (n= 2)
4– 5	6	6	25	
5– 6	5	2	12	

(Table 1). Their daily nectar and pollen foraging activity was at its maximum between 9.00 and 11.00 a.m., E.S.T. Between noon and 2.00 p.m. the number of honeybees observed on the plots decreased rapidly. After 2.00 p.m. only an occasional honeybee was observed on the observation stations. Three apiaries of some 40 colonies each were located ⅓, ⅔ and ¾ miles from the experimental safflower field. Ninety-five per cent of the native bees were bumblebees; the remaining bees were solitary bees. Most visitors other than bees were syrphid flies. Aside from considerable fluctuations in the counts, it was noted that bumblebees commenced to forage at somewhat earlier hours than honeybees. Bumblebees generally collected pollen and nectar simultaneously, while only seven out of ten honeybees observed at the same time were carrying pollen loads. It remained unknown, however, whether honeybees collecting nectar only on safflower were less effective pollinators than pollen gatherers.

The reason for the remarkable variation during the day in the magnitude of honeybee foraging activity on safflower is undoubtedly very complex. It is not fully understood at this time, albeit a number of general theories have been advanced to explain this differential which has already been observed on other crops (1, 4). It is now generally conceded that a variation in honeybee abundance is caused by a change in relative attractiveness of a plant to the bees. If this is true, the attractiveness of safflower relative to other crops blooming at the same time must have been greatest in the morning and decreased as the day progressed.

Adjoining the north and south sides of the safflower field were plantings of flax. The flax blossoms were open in the mornings only, and hence could not support bees during the afternoons. Fields of buckwheat in bloom in the vicinity and sparse growth of volunteer legumes may have successfully "competed" with the safflower in attracting the available honeybee population. In these crops the nectar and pollen productions show considerable diurnal variations, and a production increase in one crop may have adverse

TABLE 2.—SAFFLOWER SEED YIELDS FROM THREE PAIRS OF PLOTS, OTTAWA 1958

Area	Plot size (feet)	Treatment	Seed (grams)
A	6 x 6	Open	610
	6 x 6	Caged	210
B	6 x 6	Open	668
	6 x 6	Caged	320
C	6 x 12	Open	960
	6 x 12	Caged	522

effects on the attractiveness of any other crop whose productions did not, at the same time, increase in similar proportions.

The variation in the relative attractiveness of safflower to honeybees may also have been caused, in part, by changes in its own nectar and pollen yields. The honeybee population on safflower was at its peak when the nectar sugar concentration was highest, but the rapid decline in population during the early afternoon could not be attributed to nectar sugar concentration since it remained high (Table 1). To measure the amount of available nectar and pollen was not feasible, but subjective estimations indicated that during the morning visitation peak the volume of available nectar and the supply of collectable pollen decreased rapidly. By noon each day, much of the pollen which became available during the preceding night and morning, and most of the accumulated nectar, had been virtually all collected. Thereafter, only currently maturing pollen and nectar just secreted could be collected. The quantities thus available apparently sufficed for syrphids but were insufficient for honeybees to continue foraging.

There was a rather rapid cessation of flowering around August 25, and by September 1 the safflower at the experimental field was practically out of bloom except in the caged plots where blooming was prolonged by about 1 week.

The weights of seed harvested from the seed plots are tabulated in Table 2. The open pollinated plots averaged an equivalent of about 1,500 pounds of clean seed per acre, while those plots from which insects were excluded during the blooming period yielded only an average equivalent to 700 pounds of clean seed per acre. From these results it appears that insects, particularly honeybees, visit the safflower, assist in pollination, and effect increased seed yields.

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EVAPOTRANSPIRATION AT KAPUSKASING, ONTARIO

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ABSTRACT

Evapotranspiration from timothy, kept clipped and growing on moist soil and in similar surroundings, was measured during five summers at Kapuskasing, Ontario. The measured evapotranspiration was greater than evaporation from open water in a sunken evaporating pan and less than "potential" values computed on a monthly basis by the Thornthwaite and Penman formulae.

In describing the climate of any area, one of the main considerations is the moisture balance. Precipitation records are more meaningful when compared with the amount of water evaporated from crops when soil moisture is abundant. Potential evapotranspiration may be computed from weather records by formulae such as those of Thornthwaite (9) and Penman (6) but their use in any region without being checked against actual measurements is hardly justified.

In southern Ontario the water loss from turf growing on moist soil was measured at Toronto from 1947 to 1949 (7) and at Windsor in 1953 (8), while other measurements are in progress at the Ontario Agricultural College, Guelph. These places are between 41° and 43°, north latitude. In order to get similar records in northern Ontario, evapotranspiration measurements were started in 1954 at the Kapuskasing Experimental Farm. Kapuskasing is over 400 miles north of Toronto in the Clay Belt at 49°, 30′N, which is the northernmost farming section of the province. The climate is cooler and moister and the summer days longer than in southern Ontario.

EQUIPMENT AND METHODS

The installation is similar to the one used at Windsor (4) and smaller but otherwise similar to some currently in use at Guelph. It consists of a single tank, 22 inches in diameter and 30 inches deep, countersunk in the ground with a rim appearing 1½ inches above ground level. From the bottom a drain pipe 20 feet long leads into a water-tight underground chamber made from another oil drum. The tank is filled with 6 inches of gravel, then about 23 inches of sand, and topped off at ground level with a 3-inch layer of timothy sod cut from the field at the site where the soil is a clay. The setting was on high ground in a field of timothy. The tank was set in an area 50 feet square which was watered when required to keep the soil moist and this area, including the tank, was clipped regularly to a uniform height. It was bordered on the south by a hedge but in other directions there were no trees or buildings to interfere with air movements. The timothy in the tank did not form a very dense turf but maintained a healthy growth throughout.

The soil in the tank was irrigated every morning, the amount sprinkled on being enough to produce a little percolation. The previous day's excess

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TABLE 1. — EVAPOTRANSPIRATION AND RELATED VALUES, KAPUSKASING 1954-1958

Date	Meas. water use mm.	Evap. pan mm.	Th-wte P E mm.	Penman Et mm.
1954				
June	91	85	115	113
July	98	84	116	128
Aug.	79	64	95	90
Sept.	43		55	42
Oct. 1-15	18		19	13
1955				
June	119	101	131	116
July	133	119	145	135
Aug.	109	101	118	109
Sept.	58	46	61	70
Oct. 1-23	18	—	28	25
1956				
June 14-30	53		62	65
July	109	89	113	114
Aug.	78	81	104	104
Sept. 8-30	15		33	33
Oct. 1-23	19		34	28
1957				
July 17-31	54	41	64	56
Aug.	139	98	102	106
Sept. 1-26	44	33	54	52
1958				
May 13-31	56	37	39	57
June	110	71	90	115
July		73	121	124
Aug.	90	62	98	85
Sept.	52	27	71	64
Oct.	21	—	27	17

was measured at the same time. On fine days in June, July and August 0.25-inch applications were usually given although this was occasionally increased to .37 inches. In May, September and October applications of 0.12 inches usually were sufficient to provide an excess.

At the same site this Station records temperature, rainfall and humidity and maintains an evaporating pan and a black Bellani Plate atmometer. The pan is 4 feet in diameter and 2 feet deep, countersunk in the ground with a 2-inch rim showing. It is the standard type used on Canada Department of Agriculture Research Stations. The water level is kept between 2 and 4 inches below the rim. The Bellani Plate results are available for only parts of the last four seasons. They have been discussed already by Holmes and Robertson (1). Sunshine duration is recorded at the station with a Campbell-Stokes recorder, while wind speeds, humidity and solar radiation are measured also at the nearby airport where the exposure is quite similar.

Solar radiation records from the Kapuskasing airport are available from October 1954 onward, and were used in the Penman formula for evaporation from open water. Before that date the incoming radiation was estimated from sunshine records using Mateer's formula (2, 3). The comparable expression in the Penman formula gave slightly higher values for

Kapuskasing. The factor for wind used in the Penman formula was $(0.5 + \mu_2/100)$, μ_2 being 10/12 of the wind milage recorded at a height of 40 feet at the airport. In converting from open water to turf the factor of 0.8, used by Penman for summer conditions in England was applied throughout.

RESULTS AND DISCUSSION

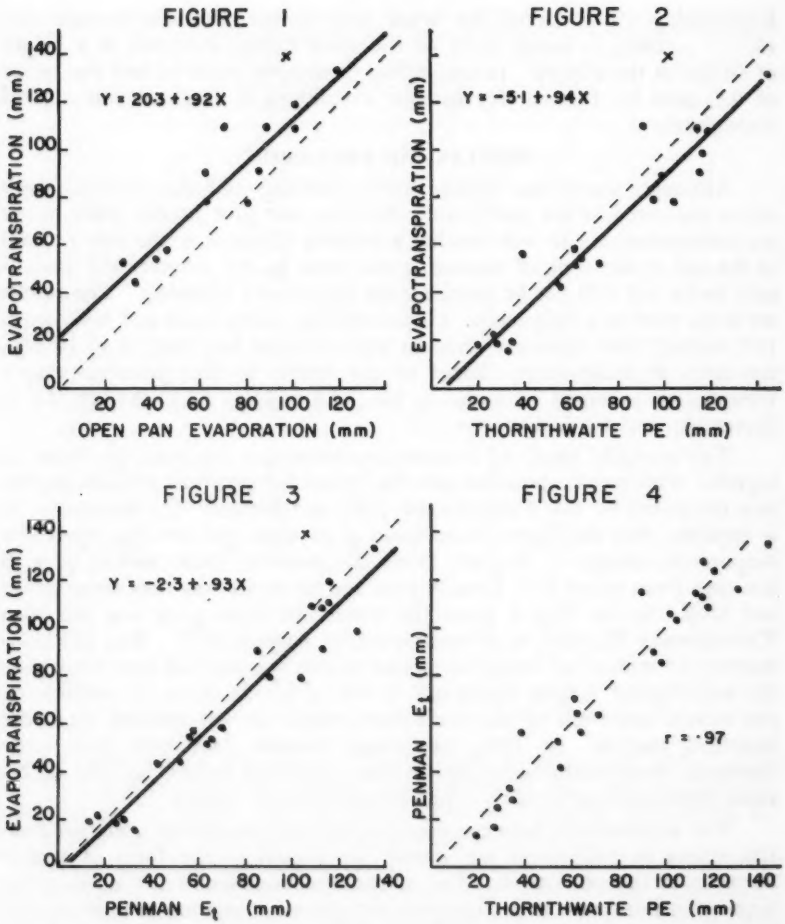
Although water was applied every morning and the previous day's excess measured at the same time, this does not give precise daily water use measurements. In wet weather a variable excess of water may be held in the soil at the time of measuring and even in dry weather the amount held in the soil will not be precisely the same every morning. The results are never used on a daily basis. However, daily water losses of 5 millimetres (0.2 inches) were common while an inch of water loss every 6 to 10 days was usual in midsummer. Based on the results to date potential evapotranspiration averaged 4.2 inches in June, 4.4 in July, 3.5 in August, 2.0 in September, and 0.9 in October.

The monthly totals of evapotranspiration are compiled in Table 1, together with pan evaporation and the values for potential evapotranspiration computed by the Thornthwaite (PE) and Penman (E_t) formulae. It is apparent that the water losses from grass were greater than open pan evaporation except in August, 1956. Evaporation from sunken pans is less than from raised U.S. Class A pans and no doubt less than from ponds and lakes. In the first 4 years the water loss from grass was less than Thornthwaite PE value in all months except August, 1957. The 139 millimetres (5.5 inches) of water loss in that month was over an inch more than the next highest August figure and is out of line in terms of evaporating pan results and both of the computed values. It was omitted from the statistical analysis. In 1958, two more months, May and June, gave measured evapotranspiration higher than computed values, but the differences were small particularly when compared to E_t values.

The relationships between measured evapotranspiration and comparative values in millimetres per month are shown in the form of scatter diagrams in Figures 1 to 4. The dotted lines represent a 1:1 relationship. Regression lines with their equations are given for relating evapotranspiration to pan evaporation, PE and E_t values, while the correlation coefficient between PE and E_t is given in the last case. August 1957, which was omitted from these calculations, is marked with a cross.

In Figure 1, monthly evapotranspiration is plotted against monthly pan evaporation and the linear regression line drawn. The evapotranspiration was greater than pan evaporation in nearly every case. The predicted value of evapotranspiration for zero pan evaporation is 20.8 millimetres. This proved to be significantly different from zero. However, the slope of the line (.92) is not significantly different from unity. Although some extrapolation is involved, this indicates that evapotranspiration from the grass starts before there is any evaporation from the sunken pan.

Figures 2 and 3 show the measured evapotranspiration plotted against computed PE and E_t and the linear regressions in the form $Y = a + bX$. In



FIGURES 1 to 4. Graphs of monthly evapotranspiration at Kapuskasing plotted against (1) Pan evaporation; (2) PE, and (3) E_t values; and (4) E_t against PE. The dotted lines represent a 1:1 relationship.

each case the predicated "a" value was not significantly different from zero and neither slope was significantly different from unity. This, however, does not imply a 1:1 relation which would require that simultaneously $a = 0$ and $b = 1$. A special test was made of this hypothesis and in each case it conflicted significantly with the data. Thus in neither case is it true to say that there is a 1:1 relationship.

The statistical tests used above are the standard ones (5, Chap. 6) "t" tests for $a = 0$ and $b = 1$, and an "F" test for the simultaneous hypothesis $a = 0$ and $b = 1$. Table 2 shows the standard errors of these tests.

TABLE 2. — STANDARD ERRORS OF SIGNIFICANCE TESTS OF REGRESSIONS RELATING MEASURED EVAPOTRANSPIRATION TO PAN EVAPORATION, THORNTHWAITE PE AND PENMAN ET

	Test 1 (a = 0)	Test 2 (b = 1)	Test 3 (a = 0, b = 1)
Meas. E. — Pan	.28*	.09NS	2.87**
Meas. E. — PE	.20NS	.06NS	1.74**
Meas. E. — Et	.16NS	.05NS	1.11**

NS—Not significant
*Significant at 5% level
**Significant at 1% level

The differences between these measured and computed evapotranspiration values are not great. However, if further measurements of potential evapotranspiration in these latitudes substantiate these results, and especially if the differences increase in more northerly latitudes, some corrections will have to be made if the climate is to be described satisfactorily.

Figure 4 is included as a matter of interest. It shows monthly Penman E_t values plotted against Thornthwaite PE. The computed monthly values by these two methods are very close, the coefficient of correlation being .97. The larger discrepancies do not appear to be due to humidity, wind or sunshine.

SUMMARY

Evapotranspiration from a timothy sward, watered daily and kept clipped was measured in a lysimeter at Kapuskasing during five summers. The measured evapotranspiration was greater than evaporation from a sunken pan of the standard type installed at agricultural research stations in Canada. The results also indicate that there was appreciable water use by turf before there was any water loss from the evaporating pan.

Monthly totals of measured evapotranspiration were significantly less than values computed according to the Thornthwaite and Penman formulae.

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STUDIES ON THE CONTROL OF DWARF BUNT IN WINTER WHEAT

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ABSTRACT

The development of satisfactory measures for the control of the dwarf bunt disease in winter wheat is complicated by the long persistence of the causal fungus in the soil and also by the occurrence of physiologic races. A brief review of literature dealing with these problems is presented.

The present paper deals primarily with the use of fungicidal chemicals in the control of this disease. Experimental results from tests conducted during the period 1954-1958 showed that chemical seed treatment was useless against soil-borne inoculum, but that liquid mercurial and chlorobenzene seed dressings were highly effective against seed-borne spores. The only substantial reduction of disease in the field was achieved by the use of chlorobenzene fungicides applied to the surface of the soil shortly after planting.

A list of recommendations for seed treatment, as a measure of limiting the spread of this disease, is given at the end of this paper.

INTRODUCTION

Early references to bunt in wheat do not distinguish between common and dwarf bunt. The first report suggesting the existence of dwarf bunt as a disease distinct from common bunt was made by Young (15) in 1935 when he described it as a new variety of *Tilletia tritici* and indicated that the inoculum was soil borne. That same year Tingey and Woodward (13), working in Utah, attributed the increasing prevalence of smut in wheat to the use of the combine harvester, which so effectively disseminated the fungus in the field. They stated that, while seed treatment was effective in destroying the smut on the grain, it could not destroy inoculum that was in the soil. For this reason they proposed the breeding of resistant varieties as the most hopeful solution to the problem. In 1936 Holton and Heald (7) observed that a race of *T. tritici*, conspicuous because of its extreme dwarfing and tillering effect on the host plant, was much more difficult to control than other races of *T. tritici* or *T. levis*. Later Bamberg (1) described the destructive nature of dwarf bunt in certain areas of Montana and observed that some of the heaviest infections occurred in fields where the seed had been treated before planting. This evidence suggested that chemical seed treatment, at least with materials in general use for the control of common bunt, was ineffective against dwarf bunt. In 1949 Holton *et al.* (6) claimed that infestations of the dwarf bunt fungus persist in soil for at least two seasons and, from observations made in disease areas where resistant varieties had been grown for a number of years, they concluded that such infestation may persist for at least 4 and possibly 7 years. The realization of this persistent nature of the inoculum stimulated the search for varietal resistance as a means of control. This search met with reasonable success in its earlier stages when Briggs and Holton (3) found that two genes, the Martin and either the Turkey or Rio, together gave resistance to all 25 known races of common bunt and also to dwarf bunt.

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Only 2 years later Holton and Vogel (9) reported heavy infections of dwarf bunt in varieties which had previously been highly resistant. They also observed that the disease occurring in these varieties differed in several respects from that previously encountered. Such evidence pointed to the presence of more than one pathogen race of the dwarf bunt fungus, a situation which immediately complicated the program of breeding for resistance.

Another approach to the control of this disease has been the use of chemical fungicides. Seed treatment with the chemicals in general use for control of common bunt appeared to be ineffective against the soil-borne dwarf bunt fungus. Attention turned to the possibility of control by applying chemicals to the soil. Possibly one of the earliest reports of such work was from Germany by Wagner (16) in 1950, when he reported that brassicol, a pentachloronitrobenzene preparation, applied at the rate of 40 grams per square metre (approximately 350 pounds per acre) largely eliminated both dwarf bunt and common bunt from the crop. Later (17) he reported that brassicol at 50 kilograms per hectare (approximately 50 pounds per acre) applied 4 weeks after sowing was completely effective against dwarf bunt. In exploratory tests Holton and Jackson (8) found that Anticarie, a chlorobenzene dust, gave significant control of dwarf bunt when applied to the soil at the rate of 100 pounds per acre. They obtained no control with standard mercurial seed dressing chemicals when these were applied to the soil in the same manner.

Studies on dwarf bunt were begun in Ontario after reports by Page (10), also Conners and Skolko (4) established its presence in the province. Field survey reports by Conners and Fushtey in 1954*, and more recently by Baylis (2), showed that the disease was present in at least 10 counties of Southern Ontario and was causing some concern among winter wheat growers, particularly those interested in the seed trade.

The purpose of the present study was to confirm results of previous workers and to obtain sufficient knowledge of the effects of different fungicides on this disease to permit an appraisal of the use of fungicidal chemicals in the control of dwarf bunt in winter wheat.

EXPERIMENTAL METHODS AND RESULTS

Chemical Seed and Soil Dressings for the Control of Soil-borne Inoculum

Experimental plots were set up in 1954 on a farm in York County where dwarf bunt was first recognized in Ontario (10). Mature bunted wheat heads found in the area were collected for use as inoculum supplementary to that already present in the soil.

Field experiments were conducted on this site in each of four seasons, namely 1954-55, 1955-56, 1956-57 and 1957-58. Disease incidence in two of these seasons, namely 1954-55 and 1956-57, was so slight (less than 1 per cent) that comparison of the various treatments gave no significant differences. Consequently, the data from these experiments are omitted and experimental details are given for only the two seasons which yielded a higher level of infection.

*Field survey for dwarf bunt in Ontario in 1954. *Unpublished mimeographed report.*

The chemicals used in these tests are described in the following table:

Designation	Description
Organic Mercurials	
Ceresan M	Dust — Du Pont of Canada Ltd.
Ceresan 200	Liquid — Du Pont of Canada Ltd.
Panogen 15	Liquid — Morton Chemicals
Puraseed	Wettable powder — Gallowhur Chemicals, Ltd.
Chlorobenzenes	
Anticarie	40% hexachlorobenzene dust — French Dyestuffs
Tritisan	60% pentachloronitrobenzene dust — Hoechst
YF308	Combination fungicide-insecticide dust containing hexachlorobenzene — Canadian Industries Ltd. (proportions unknown)
PCNB	20% pentachloronitrobenzene dust — O. Mathieson
Others	
974-90WP	Experimental fungicide-herbicide wettable powder — Carbide Chemicals Co.
Mylone	85% commercial product of above
GC1189	Experimental chlorinated wettable powder fungicide — General Chemicals
Cyanamid	44% granular calcium cyanamide — Cyanamid of Canada Ltd.
Copper sulphate	Technical grade, granular — Fisher Scientific Co., Ltd.
Formalin	40% formaldehyde — Standard Chemicals

1955-56 Experiment

The seed used for this field experiment was registered Genesee winter wheat. The plots, consisting of two 8-foot rows, replicated three times, were planted with a V-belt seeder on September 19.

Inoculum consisted of a 1:100 (by weight) spore-soil mixture that was left outdoors in a shallow heap for 3 weeks before use. Immediately after planting, this mixture was sifted through a screen and spread uniformly along the length of each row at the rate of 1 quart per row.

Ten chemicals were tested, six as seed and soil dressings and four as soil dressings only, as shown in Table 1. The seed dressings were applied in the usual manner, namely, 100-gram lots of seed were weighed into 200-millilitre erlenmeyer flasks; the required amount of chemical was carefully measured and added to the seed, and flasks were stoppered and shaken vigorously for 2 minutes to obtain thorough mixing. With liquid materials the chemical was carefully added to a glass cover slip placed on the surface of the seed prior to shaking. Soil dressings were applied as follows: Liquids, wettable powders, and soluble chemicals were added to 100 millilitres of water and sprinkled uniformly along the length of the row by means of a clothes sprinkler attachment. Dry powders were mixed with 100 cubic centimetres of white sand and spread uniformly along the length of the row.

Notes on the incidence of dwarf bunt in these plots were taken in July, 1956. Counts were taken on the total healthy as well as diseased wheat heads per row so that disease incidence could be expressed on a percentage basis. The results of this test are given in Table 1.

Statistically, most of the treatments show significantly higher or lower disease ratings than the untreated controls. However, these small differences are of no practical value. It is necessary to have a high level of disease control in order to achieve practical significance. In this regard the results in Table 1 clearly indicate that only three chemicals, YF308, Anticarie, and Tritisan, applied to the soil, gave significant disease control,

TABLE 1. — EFFECT OF SEED AND SOIL APPLICATIONS OF FUNGICIDAL CHEMICALS ON INCIDENCE OF DWARF BUNT — 1956

	Treatment		Per cent bunted heads (average 3 replicates)
YF308	Seed	2 oz. per bushel	32.9
	Soil	20 lb. per acre	7.5
	Soil	40 lb. per acre	5.1
Anticarie	Seed	1 oz. per bushel	16.3
	Soil	20 lb. per acre	5.0
	Soil	40 lb. per acre	6.3
Cer. M	Seed	1 oz. per bushel	27.3
	Soil	20 lb. per acre	24.4
	Soil	40 lb. per acre	10.0
Tritisan	Seed	1 oz. per bushel	22.6
	Soil	20 lb. per acre	6.6
	Soil	40 lb. per acre	5.9
Puraseed	Seed	1 oz. per bushel	13.7
	Soil	20 lb. per acre	20.1
	Soil	40 lb. per acre	17.1
Panogen	Seed	1 oz. per bushel	21.7
	Soil	20 lb. per acre	16.4
	Soil	40 lb. per acre	11.6
974-90WP	Soil	40 lb. per acre	53.5
	Soil	80 lb. per acre	58.4
Cyanamid	Soil	80 lb. per acre	28.4
	Soil	160 lb. per acre	30.3
$CuSo_4$	Soil	20 lb. per acre	19.7
	Soil	40 lb. per acre	15.2
Formalin	Soil	20 gal. per acre	16.3
	Soil	40 gal. per acre	11.5
Control	(No treatment)	(a)	26.0
		(b)	28.3

Standard error of the means for treatments = 1.6 per cent

each resulting in approximately 80 per cent disease reduction. It is further apparent that increasing the dosage from 20 pounds to 40 pounds per acre did not increase disease control. None of the chemicals applied as seed dressings gave significant disease control.

1957-58 Experiment

Since disease development was inadequate in two of the three previous experiments, an extra effort was made to promote the disease by using straw cover as recommended by Tyler and Jensen (14). This cover was applied in early December to the first two of the four replications.

The results of the previously described experiment, supported by personal communications with workers in the field and published reports, such as that of Holton and Jackson (8), established the fact that seed treatment was ineffective in controlling dwarf bunt in the field. Consequently, subsequent field experiments were confined to the use of chemicals as soil dressings only. The results of the 1957-58 experiment are given in Table 2.

These results confirm those of the 1956 experiment in that only the chlorobenzene materials (YF308, Anticarie, and PCNB) were effective in significantly reducing disease incidence. Straw cover increased incidence in nearly all cases with an over-all increase of approximately 50 per cent over no cover.

TABLE 2. — EFFECT OF SOIL APPLICATIONS OF FUNGICIDAL CHEMICALS ON THE INCIDENCE OF DWARF BUNT — 1958

Treatment	Per cent bunted heads			
	Rate per acre	Straw cover av. (2 rep.)	No cover av. (2 rep.)	Over-all av. for treatments
Anticarie	25 lb.	4.2	7.8	6.0
	50 lb.	7.4	0.0	3.7
PCNB	50 lb.	8.8	3.5	6.2
	100 lb.	0.0	4.4	2.2
YF 308	50 lb.	5.8	10.3	8.1
	100 lb.	19.6	0.0	9.8
Cyanamid	100 lb.	26.6	12.0	19.3
	200 lb.	17.1	14.5	15.8
Mylone	50 lb.	19.6	15.6	17.6
	100 lb.	14.3	11.1	12.7
G.C. 1189	50 lb.	21.1	11.1	16.1
	100 lb.	15.7	9.9	12.8
Cer. 200	25 lb.	21.6	8.4	15.0
	50 lb.	21.5	13.3	17.4
$CuSo_4$	25 lb.	18.9	8.1	13.5
	50 lb.	21.3	14.1	17.7
Check	(a)	26.9	21.1	24.0
(no treatment)	(b)	23.1	18.5	20.8
	Average	16.5	10.4	

Standard error of the means for treatments = 1.0 per cent

The general conclusions that may be drawn from the field tests conducted during this 4-year period are:

1. Seed treatment provides no control of this disease in the field.
2. Of all the fungicides tested as soil dressings (organic mercurials as well as organic and inorganic non-mercurials), only the chlorobenzenes gave any appreciable disease control.
3. In view of the extreme variation in disease development from one year to the next, the disease must be influenced by complex factors which cannot be established at this time. The low incidence in 1954-55 could be attributed to "Hurricane Hazel" which practically washed-out the plots in late October; but no simple explanation can be proposed for the failure in 1956-57.

Chemical Seed Dressings for the Control of Seed-borne Dwarf Bunt Spores (Laboratory Experiments)

Although seed treatment was ineffective against dwarf bunt in the field, the fact that certain chemicals were effective as soil dressings suggested that these chemicals were capable of destroying, or at least inhibiting, the inoculum of the causal fungus. If these materials were effective against dwarf bunt spores carried on wheat seed, they would be extremely useful as seed dressings in preventing the introduction of the pathogen into clean soil.

A series of tests was conducted to determine the effectiveness of a number of fungicidal seed dressings against seed-borne dwarf bunt spores. A preliminary report by the author (5) showed that Anticarie, Tritisan, as

TABLE 3. — EFFECT OF SEED TREATMENT CHEMICAL ON GERMINATION OF SEED-BORNE DWARF BUNT SPORES

Treatment	Rate per bushel	Spore germination rating			
		(2-day storage after treatment)		(7-day storage after treatment)	
		1	2	1	2
Agrox C	½ oz.	1	1	1	1
	1 oz.	1	0	1	1
Puraseed	½ oz.	1	1	1	2
	1 oz.	1	1	1	1
Ceresan M	½ oz.	2	1	1	1
	1 oz.	1	1	1	1
San	½ oz.	1	2	1	2
	1 oz.	1	1	1	0
Leytosan	½ oz.	1	2	1	1
	1 oz.	1	1	1	1
Setrete	¾ oz.	1	1	0	1
	1½ oz.	0	0	0	0
Gallotox	¾ oz.	0	0	1	0
	1½ oz.	0	0	0	0
Panogen 15	¾ oz.	0	0	0	0
	1½ oz.	0	0	0	0
Anticarie	½ oz.	0	0	0	0
	1 oz.	0	0	0	0
Tritisan	½ oz.	0	0	0	0
	1 oz.	0	0	0	0
Thioneb 50W	2 oz.	0	0	0	0
	4 oz.	0	0	0	0
Check (no treatment)		2	2	1	2

Rating symbols: 0 — no sporidia
1 — a few sporidia
2 — abundant sporidia

well as Panogen 15, prevented germination of dwarf bunt spores taken from seed that had been treated with these materials. On the other hand, spores taken from untreated seed, or that treated with Ceresan M, germinated abundantly. The methods used in the present tests were essentially the same as those described in the preliminary report (5) where the spores were incubated at 5°C. in moist soil culture and spore germination was determined on the basis of relative sporidial development rather than a numerical spore count. Table 3 gives the results of one such test conducted in May, 1956, where 11 different materials were tested at two rates of application: (a) that recommended by the manufacturer, (b) twice the recommended dosage. These results indicate the following effects:

1. The dry mercurials (Agrox C, Puraseed, Ceresan M, San, and Leytosan) did not prevent spore germination, although the number of sporidia produced was reduced in most instances.
2. The liquid mercurials (Setrete, Gallotox and Panogen 15) were much more effective, with complete control being achieved at the double dosage.

An important observation, apart from the data presented, is that the phenyl mercury's (Setrete and Gallotox) were highly phytotoxic when applied at this high dosage, causing seriously reduced germination. Panogen 15, at the same dosage, did not affect seed germination but noticeably retarded subsequent development of the shoot.

3. Chlorobenzene preparations (Anticarie and Tritisan) and Thioneb 50 (polyethylene thiuram sulphide) were fully effective at either dosage with no apparent seed injury.

In order to ascertain whether the effect of these chemicals was temporary and due to their presence in the inoculum solution, or whether the spores were rendered permanently non-viable prior to incubation, the technique of preparing inoculum was modified by including a series of spore-washing operations. This was accomplished in the following manner: Spores from 25 grams of infested, treated seed were brought into suspension by shaking in 20 millilitres of distilled water. Ten millilitres of this suspension were drawn off with a pipette, transferred to a 15-millilitre centrifuge tube, and the spores spun down. The supernatant liquid was then poured off, leaving a semi-solid mass of spores (about 0.1 millilitre volume) at the bottom of the tube. This mass of spores was re-suspended in 10 millilitres of distilled water by vigorous shaking and then spun down again. The washing operation was repeated three times, the fourth resuspension being used as the inoculum. Although no test for the presence of the fungicidal chemical in the final inoculum preparation was made, it was assumed that the chemical in solution or suspension was sufficiently dilute (approximately 1×10^{-8} of the concentration in the original preparation) that it would not likely have any inhibiting effect on spore germination.

To determine the effect, if any, of post-treatment storage on spore germination, the seed was kept in stoppered erlenmeyer flasks and sampled 1 day and again 7 days after the application of the fungicide.

The inoculum preparations were incubated in soil cultures as well as on 4 per cent water-agar test tube slopes. The 10-millilitre processed sample of inoculum was first used to inoculate two agar-culture tubes by flooding the surface and pouring back the excess. The remainder of the sample was then used to inoculate a single glass-tumbler soil-culture preparation by pouring the entire volume over the surface of the soil. Two samples were taken from each seed treatment, thus providing four agar cultures and two soil cultures for analysis. The two types of cultures were incubated side by side in a Wisconsin temperature tank held at 5°C. Readings were taken at weekly intervals by examining microscopic slide preparations. These were prepared as follows:

1. With the agar cultures, the surface of the culture was scraped lightly with a small sterile inoculating loop and the scrapings suspended in a drop of lactophenol blue on a microscope slide. A cover glass was lowered into place and the preparation examined directly for the presence of sporidia.
2. With the soil cultures, the larger soil particles presented a problem in the direct preparation of slides. Consequently, the scrapings from about 1 square centimetre of soil surface were first suspended in 1 millilitre of water in a watch glass. As soon as the larger soil particles had settled to the bottom a drop of the suspension was drawn off from the surface by means of an eye-dropper and transferred to a drop of lactophenol blue on a microscope slide. A cover glass was then put into place and the preparation examined under the microscope.

In both soil and agar cultures sporidia were first detected after 4 weeks of incubation. The results given in Table 4 were taken during the 6th week.

TABLE 4. — EFFECT OF POST-TREATMENT WASHING ON THE GERMINATION OF DWARF BUNT SPORES

Treatment	Rate per bu.	Storage period	Spore germination rating			
			Agar cultures		Soil cultures	
			washed	unwashed	washed	unwashed
Ceresan M	½ oz.	1 day	2	1	–*	–
	½ oz.	7 days	1	1	1	0
Anticarie	½ oz.	1 day	0	0	–	–
	½ oz.	7 days	0	0	0	0
Cyprex	½ oz.	1 day	2	2	–	–
	½ oz.	7 days	2	1	2	1
	1 oz.	1 day	2	2	–	–
	1 oz.	7 days	2	2	2	2
Tritisan	½ oz.	1 day	0	0	–	–
	½ oz.	7 days	0	0	0	0
Panogen	¾ oz.	1 day	1	1	–	–
	¾ oz.	7 days	0	0	0	0
Gallotox	¾ oz.	1 day	1	0	–	–
	¾ oz.	7 days	0	0	0	0
Ceresan 75	¾ oz.	1 day	0	0	–	–
	¾ oz.	7 days	0	0	0	0
Check (no treatment)		1 day	2	2	–	–
		7 days	2	2	2	2

*1-day soil cultures became accidentally flooded some time between 3 and 4 weeks' incubation. No sporidia developed in any of the treatments, so that it was considered preferable to treat this result as missing data.
Rating symbols: 0 — no sporidia
1 — a few sporidia
2 — abundant sporidia

The data in Table 4 confirmed those given in Table 3 in showing that dry mercurials were ineffective, liquid mercurials at the standard rates of application were incompletely effective, and the chlorobenzenes were completely effective in preventing sporidial development from chlamydospores of the dwarf bunt fungus. The post-treatment washing of spores apparently did not remove the inhibiting effect of the most effective fungicides. With the less effective ones there was some indication of reduced inhibition after washing, e.g., with Ceresan M ½ oz., 1-day storage, more sporidia were produced in both agar and soil cultures when the spores had been put through the washing operation. A similar result was obtained with the Cyprex ½ oz., 7-day storage treatment. The chlorobenzene materials (Anticarie and Tritisan) gave complete control regardless of post-treatment storage and washing operations. This result shows that the effect of these fungicides is permanent, i.e., it is not removed when the fungicide is removed by washing. The results also show that most of the fungicidal effect is brought about within 24 hours after treatment, this being the shortest post-treatment storage period in the test.

DISCUSSION AND CONCLUSIONS

Two characteristics of the dwarf bunt fungus are largely responsible for the difficulty in developing satisfactory control methods: 1) the persistent nature of the chlamydospores in the soil, 2) the occurrence of physiologic races. The first can be overcome in part by long-term crop rotations;

the second by the development of new resistant varieties, but neither has solved the problem of adequate control of this disease.

The use of fungicidal chemicals has become almost universal in the study and development of plant disease control measures. With dwarf bunt a high degree of control can be achieved by the use of chlorobenzene fungicides as soil dressings. This has already been demonstrated by a number of workers (8), (11), (12), (17), and others. The question arises as to whether this method is economically feasible. The lowest dosage achieving a high degree of control was that reported by Purdy (11), who showed that 10 pounds of Sanocide (40 per cent HCB) per acre applied 4 weeks after emergence reduced dwarf bunt incidence from 38 per cent in the untreated controls to 0.5 per cent in the treated plots. At this rate the cost of material for such treatment is not prohibitive but still high for a crop such as winter wheat. Furthermore, experimental results of other workers and those reported here indicate that control is seldom as highly effective as shown in the example cited above. For these reasons, as well as the fact that this would require an additional field operation, this method of control is not likely to be adopted to any large extent in Canada.

More acceptable measures for control would seem to be the introduction of resistant varieties into dwarf bunt areas and the general use of proper chemical seed treatment to prevent the introduction of the pathogen into dwarf-bunt-free soil.

There are no dwarf-bunt resistant varieties of winter wheat generally available in Canada at the present time, but recently developed varieties are being tested here and may be available in the near future.

With respect to proper chemical seed treatment, the results presented here show that liquid mercurials applied according to standard recommendations give a high degree of protection against seed-borne inoculum. Control was not always complete at the standard dosages but was complete in all cases where these dosages were doubled. Thus, a higher than standard dosage may be considered for this purpose but, in view of the risk of serious seed injury, it would be unwise to make such a recommendation.

The results given in Table 4 indicate that, with the liquid mercurials, a post-treatment storage period in excess of 1 day is necessary for best control. In this instance both Panogen and Gallotox permitted slight sporidial development with the 1-day period, but gave complete control when stored for 7 days after treatment. When these results are compared with those of Table 3 it is apparent that a 2-day post-treatment storage period is adequate as there is no increased control when this period is increased to 7 days. Chlorobenzenes were fully effective at recommended rates of application and at the minimum (1-day) post-treatment storage period used.

In conclusion, the results of these spore-germination experiments can be interpreted in terms of practical recommendations as follows:

1. Since a high degree of protection from seed-borne spores of the dwarf bunt fungus is achieved by the use of standard liquid mercurial seed dressings, it is imperative that all winter wheat seed be treated in this manner.

2. Since there is some evidence that post-treatment storage increases the effectiveness of these chemicals against these spores, the treated seed should be stored for a period of at least 48 hours before planting.
3. Since chlorobenzene seed dressings are fully effective against seed-borne dwarf bunt spores their use should be encouraged in the control of this disease. However, since chlorobenzenes, used as seed dressings, are not effective against seed rot and seedling blight it may be desirable to use them in combination with mercurial seed dressings or captan in order to provide this protection where necessary.

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SOME EFFECTS OF FOUR CHLORINATED POLYCYCLIC INSECT TOXICANTS ON THE PHYSIOLOGY OF POTATOES, CARROTS AND RADISH

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ABSTRACT

The physiological effects on certain plants of the chlorinated polycyclic insect toxicants aldrin, isodrin, dieldrin and endrin, when these were mixed with soil, were investigated. Under field conditions 3.5 and 6.5 pounds of toxicant per acre allowed maximum yields with potatoes and carrots; and at 9.5 pounds per acre yields were greater than those for the controls.

Only minor changes in carotene and ascorbic acid content were observed. The chlorine content of both test plants was increased. In radish, ascorbic acid was generally depressed by the four compounds but nitrate uptake was increased by the epoxy compounds, dieldrin and endrin. The non-oxygenated compounds, aldrin and isodrin, depressed the nitrate content. Phosphorus content was depressed at higher application rates, while at lower levels moderate increases in phosphorus content were observed in some cases.

Within certain concentration limits, all four compounds stimulated germination of radish seed in soil. On agar media, 40 p.p.m. aldrin inhibited germination of radish seed, while 40 to 200 p.p.m. of dieldrin, endrin and isodrin stimulated germination.

The presence of the epoxide ring appeared to be associated with physiological activity whereas, with one exception, the stereo-chemical configuration of the compounds was not associated with their activity.

INTRODUCTION

Since the release of DDT (dichlorodiphenyltrichloroethane) as an insecticide, a large number of insecticidally potent organic compounds have been synthesized. These organic compounds have a much wider range of effectiveness than the older inorganic insecticides. Aldrin, dieldrin, isodrin (isomer of aldrin) and endrin (isomer of dieldrin) are cyclic chlorinated Diels-Alder condensation compounds which are insecticidally potent and in many instances more effective than DDT or some of the older organic insecticides.

Due to the high insecticidal potency of organic insecticides such as DDT, toxaphene, parathion, chlordane and the compounds used in the experiments described herein, the low but effective dosage rates of these types of compounds do not, as a rule, cause gross morphological effects on common crop plants (3, 4, 8). Some lesser but nonetheless interesting effects that these compounds may exert have been observed.

Randall (16) observed that chlordane, DDT and benzene hexachloride (BHC) stimulated the germination of red clover seeds at certain dosage levels. Significant growth and yield differences were also observed. The size and distribution of nodules were affected, whereas nitrogen and phosphorus levels in the foliage appeared to be unaffected. Bioassay of the clover plants failed to reveal evidence of absorption of the insecticides. Similar results have been obtained elsewhere. Investigations at Kansas State

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College (1) indicated that no aldrin residues were present in potatoes and tomatoes using soil applications up to 5 pounds of actual compound per acre. Cabbage, onion and sweet corn grown in soil treated with 100 pounds of aldrin per acre (this is 20 to 50 times the amount required for economical insect control) apparently contained no residual compound.

The failure to detect residues of insecticidally active organic compounds in plant tissue was probably due to the lack of sufficiently sensitive and specific analytical methods. The investigations of Lichtenstein (15) show that crops grown in soils treated with aldrin contained within their tissue both aldrin and dieldrin. Carrots were shown to absorb more insecticide than beets, cucumbers, potatoes, radishes and rutabagas. The occurrence of dieldrin in the tissues when only aldrin was applied can be attributed to the epoxidation of the olefinic double bond in aldrin to yield the corresponding oxygenated compound. Gannon and his associates (10, 11) have shown that heptachlor and aldrin are oxidized in the soil and in plants to the corresponding epoxy compounds.

No phytotoxic reactions were observed by Kuitert and Tissot (14) when dieldrin was used to control insect pests on tobacco. In contrast, Crowell and Morrison (5) reported that aldrin and dieldrin, as well as some other similar insecticides, had phytotoxic effects on cucurbits if the compounds were applied under relatively moist conditions. Squash varieties of the species *Cucurbita maxima* appeared to be more tolerant than other varieties. Phytotoxic effects of some chlorinated insecticides have also been observed by Cullinen (6). Foster (9) reported that a variety of different effects on plants were observed when using aldrin, dieldrin and other insecticides in the soil to combat soil-borne insect pests. Richardson (17) observed that dieldrin caused some stimulation of barley seedling development and that aldrin and endrin exerted an indirect effect by reducing seedling infection by a species of *Helminthosporium* while isodrin showed no effect.

Organoleptic tests gave the first indications that the organic insecticides could be absorbed by plants. The general observations (12, 13) on the induction of off-flavours suggest that the compounds may become systemic either in their original chemical form or possibly as metabolic altered derivatives.

The experiments reported in this paper concern the investigation of some physiological effects that these organic insecticides have on potato, carrot, and radish plants.

METHODS

Field Experiments:

(a) *The Effect of Isodrin and Endrin on Carrots*

Four rates of application of the compounds were used: 0.5, 3.5, 6.5 and 9.5 pounds of actual isodrin and endrin per acre and control plots where no insecticide was used.

Treatments were replicated nine times in a randomized block plot design. The usual cultural practices were carried out during the growing

season. The harvested roots, which were not stored longer than 3 weeks, were sectioned and ground in a vegetable grinder. Samples (5 grams), in duplicate, from each plot replicate were transferred to jars, layered with acetone and the containers tightly sealed for future carotene estimations. Duplicate samples (10 grams) of the ground tissue were placed in 40 millilitres of *N* NaOH for chlorine determinations. Similarly samples (20 grams), in duplicate, from each replicate were dried to constant weight in an electric drying oven to yield dry matter values. The dried samples were later ashed in an electric muffle furnace.

The method used for carotene estimation was that developed by Booth (2). The standard solution for comparison was a 0.02 per cent potassium dichromate solution which was equivalent to 30 milligrams of carotenoid as determined by a check against a sample of pure beta-carotene.

For the determination of chlorine, the alkali suspension of the tissue was boiled on a hot plate until almost dry. The residue was suspended in water and boiled for 1 hour, after which the solution was acidified with nitric acid, the volume adjusted and the solution decolorized with carbon. To an aliquot of the clear filtrate was added an excess of silver nitrate solution followed by titration with a standardized solution of potassium thiocyanate.

(b) *Effect of Aldrin and Dieldrin on Potatoes*

A similar plot design and identical rates of application of the compounds were used as for the field experiment with carrots. The usual cultural practices were employed during the growing season. The tubers were harvested when mature and 25 tubers were selected, at random, from each replicate and stored in a suitable storage compartment not longer than 4 weeks. For analyses, cross-sections of the tubers were cut, ground in a vegetable grinder and samples (5 grams) of the freshly ground material were quickly covered with 25 millilitres of 0.4 per cent solution of oxalic acid. Ascorbic acid determinations were performed on these samples. Duplicate samples (10 grams) were covered with 40 millitres of *N* NaOH for chlorine determinations. Samples (20 grams), in duplicate, of freshly ground material were dried in an electric drying oven to constant weight for dry matter values. The dry samples were later ashed in an electric muffle furnace.

Chlorine analyses were carried out as described previously. Ascorbic acid was determined by the method of Robinson and Stotz (18).

Effect of the Compounds on Germination, Growth and Chemical Composition of Radish:

(a) *Germination of radish seeds on bacto-agar containing various concentrations of the compounds*

To agar solutions were added the compounds dissolved in acetone and the mixtures boiled to expel the acetone. The agar solutions were made to volume with hot water and the preparations, in which the compounds were present in the form of a very fine suspension, then transferred to sterile petri plates. Concentrations of the compounds were 40, 200 and

600 p.p.m. and each treatment and the control were replicated three times. In each plate were placed 20 seeds and germination counts were made at 48-, 72-, 96-, 120-, 144- and 168-hour intervals.

(b) *The effect of soil applications of the compounds on germination, growth and nutrient uptake of radish plants*

Radish plants were grown in soil to which the compounds had been added at the rate of 0, 1, 5, 10, 20, 40 and 80 pounds per acre. In each flat 32 seeds were planted and each treatment was replicated three times. The flats were kept in a greenhouse until all viable seeds had germinated, after which they were transferred outdoors. Germination counts were made 3, 4, 5 and 6 days after planting and a final count made before the plants were harvested. Samples (5 grams) of freshly ground root tissue were covered with 25 millilitres of 0.4 per cent oxalic acid for ascorbic acid determinations. Nitrate nitrogen and phosphorus in leaves and in roots were determined colorimetrically, using a Klett-Summerson colorimeter. Standard calibration curves using reagent grade KNO_3 and Na_2PO_4 were constructed. In both cases, a linear relation was obtained when plotting absorbency against concentration.

RESULTS AND DISCUSSION

Foliar and root growth of carrots under field conditions in the presence of various concentrations of isodrin and endrin (Table 1) show that moderate growth stimulation appears between application rates of 3.5 to 6.5 pounds of compound per acre. A similar trend is evident in the experiment with potatoes using soil applications of aldrin and dieldrin (Table 2). With the above two crops, under the conditions of the experiments, the insecticidally potent compounds appeared to stimulate growth within certain concentration limits. This effect was noticeable at the lower application rates which, incidentally, were still in excess of the rates suggested for insect control.

TABLE 1.—AVERAGE YIELD OF ROOTS AND FOLIAGE AND AVERAGE CHLORINE AND CAROTENE CONTENT OF FRESH ROOTS OF CARROTS GROWN IN SOIL TREATED AT FOUR RATES OF ISODRIN AND ENDRIN

Compound	Rate of application (lb./acre)	Yield (oz.) Roots	Foliage	Chlorine (roots) (mg./100g. F.W.)	Carotene (roots) (mg./100g. F.W.)
—	Control	1173	296	78.6	3.86
Isodrin	0.5	1264	330	80.3	3.57
Endrin	0.5	1312	322	83.0	3.50
Isodrin	3.5	1318	324	82.2	3.46
Endrin	3.5	1418	349	84.4	3.17
Isodrin	6.5	1323	320	87.1	3.66
Endrin	6.5	1322	337	89.5	3.35
Isodrin	9.5	1288	334	88.9	3.64
Endrin	9.5	1267	316	89.8	2.96
L.S.D. .05	—	138	34	8.9	0.44

Since the insecticide compounds are highly chlorinated, chlorine determinations have been used as a measure of the concentration of these compounds in residues. Determination of chlorides by the method described previously does not show whether the chlorine originates from the intact compound in the tissue or from some metabolically altered form of

TABLE 2.—AVERAGE YIELD, CHLORINE AND ASCORBIC ACID CONTENT OF POTATO TUBERS GROWN IN SOIL TREATED AT FOUR RATES OF ALDRIN, AND DIELDRIN
(Figures represent means of nine replicates)

Compound	Rate of application (lb./acre)	Tuber yield (oz.)	Chlorine (mg./100g. F.W.)	Ascorbic acid (mg./100g. F.W.)
	Control	807	21.5	13.7
Aldrin	0.5	905	38.9	15.1
Dieldrin	0.5	851	39.0	15.8
Aldrin	3.5	1011	41.3	15.9
Dieldrin	3.5	963	41.5	14.7
Aldrin	6.5	1026	48.0	15.4
Dieldrin	6.5	904	44.8	12.6
Aldrin	9.5	960	48.8	13.8
Dieldrin	9.5	864	45.3	14.1
L.S.D. .05	—	126	6.3	1.4

TABLE 3.—AVERAGE YIELD OF ROOTS AND FOLIAGE AND THE ASCORBIC ACID CONTENT OF ROOTS OF RADISH GROWN IN SOIL TREATED AT SIX RATES OF ALDRIN, ISODRIN, DIELDRIN, AND ENDRIN
(Figures represent means of three replicates)

Compound	Application rate (lb./acre)	Yield (g.)		Ascorbic acid (mg./100g. F.W.)
		Roots	Foliage	
—	Control	110	163	15.4
Aldrin	1	123	161	15.4
	5	118	129	13.8
	10	121	137	12.5
	20	131	137	11.9
	40	140	138	10.9
	80	86	109	11.6
Isodrin	1	77	131	11.3
	5	105	147	13.8
	10	90	121	11.7
	20	109	128	9.6
	40	115	152	8.8
	80	99	127	7.5
Dieldrin	1	169	206	13.8
	5	147	254	12.5
	10	131	199	12.5
	20	133	242	12.5
	40	131	232	8.3
	80	183	186	6.3
Endrin	1	169	187	12.5
	5	155	216	8.8
	10	124	202	7.5
	20	123	214	8.4
	40	132	201	8.4
	80	96	191	8.8
L.S.D. .05	—	16	22	1.4

the original compound. Ionic chloride is also measured by the method. Colorimetric methods have been developed by Danish *et al.* (7) which are more or less specific for the compounds. Recently, a new method based on reactions of the epoxide ring have been reported for dieldrin and endrin by Skerrett and Baker (19). Bioassay results reported by a number of investigators suggest that no intact compound is translocated as such. However, examination of the results of chlorine determinations presented (Tables 1 and 2) reveal that the chlorine content of carrot roots and particularly potato tubers was increased. Based on bioassay results reported by others, it appears that the compounds may either be dehalogenated in the plant or transported as metabolically altered forms.

Isodrin and endrin appeared to have a depressing effect on the carotene content of carrots (Table 1) and aldrin and dieldrin increased the concentration of ascorbic acid in potato tubers (Table 2). It is doubtful, however, if the compounds exert any appreciable effect in the carotene biosynthetic system, nor does it appear probable that they act in some manner in biochemical reactions involving ascorbate.

The effect of the compounds on the growth of radish can to some extent be correlated with the chemical structures of the compounds. Thus the two compounds containing an epoxy ring (dieldrin and endrin), which may be a point of metabolic attack, with one isolated exception (80 pounds endrin/acre, Table 3) stimulated growth. The two non-oxygenated compounds, aldrin and isodrin, depressed foliar growth but exerted little or no effect on root development. It appears that, although the slight chemical modification of the introduction of the epoxide ring at positions 6 and 7 has little effect on the insecticidal potency with certain plants and within certain concentration ranges the epoxy compounds stimulated growth, whereas the nonoxygenated compounds were either ineffective or reduced growth at higher concentrations. The stereochemical conformation of the compounds did not appear to play a role since no major differences were evident between the endo-endo and end-exo isomers of the two related compounds.

All compounds reduced the ascorbic acid content in radish (Table 3). Since the biosynthetic pathway of ascorbic acid in plants is relatively well defined, it is interesting that in the radish plant a sensitive system was apparently encountered.

Interference in the absorption and transport of certain plant nutrients can usually be easily detected visually. Nitrogen and phosphorus deficiencies can be detected as well defined deficiency symptoms. Radish, particularly when grown in the presence of high concentrations of the non-oxygenated compounds, exhibited severe nitrogen deficiencies. In contrast, the epoxy compounds induced foliage with a dark green colour. The visual observations were borne out by the results of nitrate-nitrogen determinations (Table 4). It appeared that the assimilation and/or the deficiency of nitrate nitrogen was a function of the presence, or absence, of the epoxide ring. The epoxy compounds, dieldrin and endrin, increased nitrate uptake whereas the non-oxygenated compounds, aldrin and dieldrin, reduced it.

TABLE 4.—AVERAGE NITRATE AND PHOSPHORUS CONTENT OF ROOTS AND FOLIAGE OF RADISH GROWN IN SOIL TREATED AT SIX RATES OF ALDRIN, ISODRIN, DIELDRIN AND ENDRIN
(Figures represent means of three replicates)

Compound	Application rate (lb./acre)	Nitrate (p.p.m.)		Phosphorus (p.p.m.)	
		Roots	Foliage	Roots	Foliage
—	Control	186	164	47	182
Aldrin	1	65	72	64	163
	5	71	57	46	112
	10	65	59	39	80
	20	69	51	40	92
	40	80	44	46	102
	80	77	35	30	96
Isodrin	1	126	88	62	175
	5	86	73	38	145
	10	111	55	47	114
	20	69	45	30	113
	40	76	43	33	109
	80	92	8	32	98
Dieldrin	1	206	320	51	247
	5	224	346	48	173
	10	240	413	54	133
	20	213	460	48	189
	40	186	400	33	166
	80	226	226	28	173
Endrin	1	173	171	50	174
	5	153	156	36	161
	10	197	173	39	153
	20	124	276	42	108
	40	166	373	37	163
	80	160	293	36	109
L.S.D. .05	—	18	20	6	17

Except at lower application rates, where a slight increase in phosphorus content was observed in some cases, a moderate reduction in phosphorus content was generally evident (Table 4). There did not appear to be an association between the decrease of phosphorus content in the tissue and the chemical configuration of the two classes of compounds.

When the compounds were incorporated into the soil (Table 5), with few exceptions, germination of radish seeds was hastened and the total number of seeds that germinated was increased. The non-oxygenated compounds (aldrin and isodrin) appeared to retard growth after the germination process had taken place. The stimulation of germination may be in some way associated with an induced higher porosity of the seed coats as a consequence of the high solubility of the compounds in non-polar media. Thus the compounds might be expected to have an affinity for the lipid constituents in the seed coat, thereby altering permeability.

Since in the soil system chemical interactions in the germination of seeds may be relatively complex, the use of synthetic media in which various concentrations of the compounds were included would result in a more simplified analysis of the effects that the compounds may exert. Such experiments revealed that aldrin completely inhibited germination of radish seed on treated agar media at concentrations at which the other compounds

TABLE 5.—GERMINATION OF RADISH SEEDS GROWN IN SOIL TREATED AT SIX RATES OF ALDRIN, ISODRIN, DIELDRIN AND ENDRIN

Compound	Lb. actual compd./acre	Number[1] of seeds germinated in:				Final count
		5 days	4 days	5 days	6 days	
Control	0	0	28	48	55	65
Aldrin	1	1	43	62	66	71
	5	6	46	60	67	74
	10	0	41	60	66	73
	20	5	40	59	60	70
	40	4	32	58	59	71
	80	0	39	58	67	71
Isodrin	1	3	35	55	63	65
	5	2	52	63	68	75
	10	2	43	56	65	73
	20	1	43	61	69	76
	40	37	46	63	69	74
	80	12	41	57	65	71
Dieldrin	1	0	18	37	49	71
	5	1	30	56	66	75
	10	0	31	51	61	65
	20	0	17	50	62	75
	40	0	20	49	56	66
	80	0	18	44	54	66
Endrin	1	0	11	36	55	66
	5	0	21	51	68	77
	10	2	33	51	66	77
	20	3	39	53	58	65
	40	0	27	41	56	64
	80	0	29	47	55	58

[1]Total number possible 96

TABLE 6.—GERMINATION OF RADISH SEEDS GROWN ON AGAR CONTAINING VARIOUS CONCENTRATIONS OF ALDRIN, ISODRIN, DIELDRIN AND ENDRIN

Compound	Concentration (p.p.m.)	Total number of seeds germinated of possible 60					
		48 hr.	72 hr.	96 hr.	120 hr.	144 hr.	168 hr.
Control	0	8	33	37	45	47	47
Dieldrin	40	20	54	56	57	57	57
	200	12	39	49	51	55	55
	600	0	28	33	37	41	43
Endrin	40	16	45	53	54	54	54
	200	11	38	41	47	50	50
	600	2	26	35	41	44	44
Isodrin	40	18	49	53	53	53	53
	200	12	45	54	54	54	54
	600	0	6	20	32	37	40
Aldrin	10	10	33	46	47	50	50
	20	3	17	31	41	44	50
	40	0	7	14	21	27	30
	200	0	0	0	0	0	0
	600	0	0	0	0	0	0

either stimulated germination or had only a moderately inhibitory effect (Table 6). Both the epoxy compounds increased germination, particularly at the lower concentrations. The two stereochemically related, non-oxygenated compounds, however, had more diverse effects. Since their molecular composition is identical, the difference in their activity may be related to their stereochemical configuration.

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THE INHERITANCE OF RUST RESISTANCE

VII. THE INHERITANCE OF RESISTANCE TO RACES 15B AND 56 OF STEM RUST IN ELEVEN COMMON WHEAT VARIETIES OF DIVERSE ORIGIN[1]

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ABSTRACT

In an attempt to locate new genes for stem rust resistance, 11 varieties of wheat of as diverse origins as possible were studied. Each variety was backcrossed to the susceptible parent Marquis. The F_2 families from the backcrosses were tested with races 15B and 56, and the inheritance of resistance determined. The genes carried by each variety were identified from the results of test crosses with varieties carrying known genes and with lines of Marquis carrying single genes for resistance. Most of the resistance present in the 11 varieties could be accounted for on the basis of known genes (*Sr6* – *Sr10*). However, at least one new gene conditioning moderate resistance to race 15B, and possibly one or more new genes conditioning moderate resistance to race 56, were identified.

INTRODUCTION

In the first three papers of this series Knott and Anderson (5) and Knott (3, 4) described the inheritance of resistance to races 15B and 56 of stem rust in 20 varieties of spring wheat. A surprising result of these studies was that only 7 genes for resistance were identified and only 3 conditioned resistance to race 15B. Green *et al.* (2) have shown that these genes can account for the reaction of the 20 varieties to a large number (103 cultures) of North American races of rust. In only two cases did it appear that a variety carried genes for resistance other than those previously identified.

Since rust races are constantly changing in prevalence and new races often arise, identification of a considerable number of genes for resistance is desirable. These genes can then act as a store to be drawn on as new sources of resistance are needed in wheat breeding programs. With this in mind, 11 varieties of as diverse origin as possible were selected for study. It was hoped that the group would contain genes for resistance not already identified.

MATERIALS AND METHODS

Seed of the 11 varieties described below was obtained from the Plant Industry Station, United States Department of Agriculture, Beltsville, Maryland.

Kenya 122.D.I.T.(L) (C.I.12186), Kenya N.B.263 J (L) (P.I.117526), Kenya 341 0.2.B.I. (P.I.177183) and Kenya 318 A.J.4.A.1 (C.I.12881) came from Kenya. Each of them has good resistance to races 15B and 56 at moderate temperatures.

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No. 466-4-M-M-M (P.I.159098) originated in South Africa. It has high resistance to races 15B and 56 at moderate temperatures.

P.I.60599 is an introduction from Abyssinia and has good resistance to both races.

Sapporo haru Komugi ichigo (P.I.81790-2) is a variety of Japanese origin. It has good resistance to race 15B but is moderately susceptible to race 56.

Magnif G (P.I.197663) was produced in Argentina from the cross (H44-Sinvalocho MA) X Brazilian selection 2018/37 (from Fronteira X Mentana). It has moderate resistance to races 15B and 56.

Frontana (C.I.12470) and Rio Negro (C.I.12469) both originated in Brazil. Frontana is a selection from the cross Fronteira X Mentana. Frontana and Rio Negro are moderately resistant to races 15B and 56.

Kentana 52 (C.I.13085) was produced in Mexico. It is a selection from Kentana 48 which came from the cross Kenya C9906 X Mentana. It has good resistance to races 15B and 56.

Each variety was crossed to the susceptible parent Marquis and the F_1 plants backcrossed to Marquis. Between 75 and 100 seeds were obtained from each backcross but unfortunately a late spring frost and some wireworm damage reduced the stand of backcross plants to about 50 per cent. Consequently, the number of F_2 families tested from each backcross was smaller than intended.

In previous studies, extensive use had been made of diallel crosses to identify the genes carried by different varieties. The development by Knott [see Green *et al.* (2)] of a series of Marquis lines each carrying one of the genes *Sr6* to *Sr10* inclusive has resulted in a simplification in procedure. In the present study a small F_2 population from each of the 11 crosses with Marquis was tested for seedling reaction to races 15B and 56. The results gave a preliminary indication of the number of genes carried by each variety and the type of reaction conditioned by them. On the basis of these results, the parents were crossed, either to the appropriate Marquis lines carrying single genes for resistance, or to certain varieties of known genotype.

The F_2 plants from the crosses and backcrosses to Marquis were tested in the greenhouse for seedling resistance to races 15B and 56 at moderate temperatures. The F_2 plants from test crosses were infected with either or both of the races depending on whether the gene in question gave resistance to both or only to one race.

The procedures used in conducting seedling rust tests have been described by Knott and Anderson (5).

RESULTS

The rust reactions of the parents and of lines and varieties used as genetic testers in these studies are given in Table 1.

The actual segregations obtained in crosses and backcrosses involving the 11 parent varieties will be discussed either for each variety separately

TABLE 1. — RUST REACTION OF THE PARENTS AND OF LINES AND VARIETIES USED AS TESTERS

Variety	Seedling reaction — race 56 (pustule type)	Seedling reaction — race 15B (pustule type)	Field reaction — race 15B (per cent rust)
Marquis	4	4	40 — 60
Kenya 122	0;	0;	2 — 10
Kenya N.B. 263	0;	0;	0 — T
Kenya 318	0;	0;	0 — T
Kenya 341	0;	0;	0 — T
No. 466	0;	0;	0 — T
P.I. 60599	0; — 1⁻	0; — 1⁻	0 — T
Sapporo	3	0; — 1⁻	10 — 20
Magnif G	2⁻	2⁻ — 2	20 — 30
Frontana	2⁻	2⁻ — 2	10 — 30
Rio Negro	2⁻	2	5 — 10
Kentana 52	0;	0;	T — 5
Kenya 58 — Marquis[6] (*Sr6*)	0;	0;	
Egypt Na101 — Marquis[6] (*Sr7*)	4	1⁺ — 3⁻	
Red Egyptian — Marquis[6] (*Sr8*)	2	2	
Kenya 117A — Marquis[6] (*Sr9*)	2	4	
Egypt Na95 — Marquis[4] (*Sr10*)	2⁺ — 3	4	
Egypt Na95 (*Sr7, Sr9, Sr10*)	1⁺ — 2	1⁻ — 1⁺	
Egypt Na101 (*Sr7*)	3+	1+	

TABLE 2. — RESULTS OF SEEDLING TESTS ON F_2 FAMILIES FROM THE BACKCROSS OF KENYA 318 TO MARQUIS

		Race 15B		Totals (race 56)	Expected (1:1)	P
		Number of families				
		Seg. 1VR:3S	S			
Race 56	Seg. 3VR:1S	28	0	28[1]	24.5	.30 — .50
	S	0	21	21	24.5	
	Totals (race 15B)	28[2]	21	49	49	
	Expected (1:1)	24.5	24.5	49		
	P	.30 — .50				

The ratios within segregating families were as follows:
Race 56
[1]368VR:144S plants — P for 3:1 ratio = .10-.20
Race 15B
[2]132VR:411S plants — P for a 1:3 ratio = .70-.80

or for groups of similar varieties. In calculating the expected ratios it has been assumed that *Sr6* is dominant with race 56 but recessive with race 15B and that *Sr7*, *Sr8* and *Sr9* are dominant and *Sr10* is recessive. With both race 15B and race 56, *Sr6* normally conditions a hypersensitive reaction.

TABLE 3. — RESULTS OF SEEDLING TESTS ON F_2 FAMILIES FROM THE BACKCROSSES OF KENYA 122, KENYA N.B.263 AND KENYA 341 TO MARQUIS

		Race 15B Number of families		Totals (race 56)	P (2:1:1)	Ratios within segregating families
		Seg. 1VR:3S	S			
Kenya 122-Marquis² F_2						
	Seg. 3R:1S or 13R:3S	22	0	22		610R:181S – P(25:7) = .30-.50 (assuming half 3:1 and half 13:3)
Race 56	Seg. 1MR:3S	0	7	7	.50.-.70	38MR:123S – P(1:3) = .50-.90
	S	0	8	8		
	Totals (race 15B)	22*	15	37		*204VR:588S – P(1:3) = .50-.90
	P (1:1)	.10-.30				
Kenya N.B. 263-Marquis² F_2						
	Seg. 3R:1S or 13R:3S	20	0	20		1054R:301S – P(25:7) = .90-.95 (assuming half 3:1 and half 13:3)
Race 56	Seg. 1MR:3S	0	10	10	.30-.50	124MR:281S – P(1:3) = <.01 (the poor fit is largely due to 2 of 10 families)
	S	0	5	5		
	Totals (race 15B)	20**	15	35		**264VR:696S – P(1:3) = .05-.10
	P(1:1)	.30-.50				
Kenya 341-Marquis² F_2						
	Seg. 3R:1S or 13R:3S	24	0	24		829R:222S – P(25:7) = .50 – .90 (assuming half 3:1 and half 13:1)
Race 56	Seg. 1MR:3S	0	17	17	.30-.50	111MR:337S – P(1:3) = >.99 (only tests giving clear segregations are included)
	S	0	10	10		
	Totals (race 15B)	24***	27	51		***230VR:654S – P(1:3) = .30-.50
	P (1:1)	.50-.90				

The gene *Sr7* conditions a type 1-1⁺ reaction and causes a distinctive yellowing particularly at the tips of the leaves. Plants carrying *Sr8* give a typical type 2 reaction to both races while plants with *Sr9* give a type 2 reaction only to race 56. The line carrying *Sr10* should normally show a type 2+ reaction to race 56 but it tended to be more susceptible in these tests.

Kenya 318

The results of seedling tests with races 15B and 56 on F_2 families from the backcross of Kenya 318 to Marquis are given in Table 2. About half the families segregated for a highly resistant reaction to both races and it is clear that one gene is segregating. A cross was made with Kenya 58-Marquis[6] (*Sr6*). All of the 169 F_2 plants tested gave a fleck reaction to races 15B and 56, showing that the gene present in Kenya 318 is *Sr6*.

Kenya 122, Kenya N.B. 263 and Kenya 341

Kenya 122, Kenya N.B. 263 and Kenya 341 behaved very similarly and will be considered together. The data obtained from backcrosses involving the three varieties are given in Table 3. In each case approximately half the families segregated for a very resistant (hypersensitive) reaction to both 15B and 56. It is evident that each variety carries one gene which conditions a fleck reaction to both races. In addition, about half the families which did not segregate for a fleck reaction to both races did segregate for moderate resistance to race 56. The results show that a second gene conditioning moderate resistance to race 56, is present in each variety.

TABLE 4. — RESULTS OF SEEDLING RUST TESTS ON F_2 POPULATIONS FROM TEST CROSSES INVOLVING KENYA 122, KENYA N.B.263 AND KENYA 341

Cross	Race	Number of plants			Expected	P
		VR	MR	S		
Kenya 122						
X Kenya 58 — Mar.[6] (*Sr6*)	15B	120		0		
	56	120		0		
X Kenya 117A — Mar.[6] (*Sr9*)	56	25		3	61:3	.20 — .30
X Egypt Na95 — Mar.[4] (*Sr10*)	56	107		22	55:9	.30 — .50
X Rio Negro (*Sr8, Sr9*)	56	81		1	253:3	1.0
Kenya N.B.263						
X Kenya 58 — Mar.[6] (*Sr6*)	15B	139		0		
	56	191		0		
X Red Egyptian — Mar.[6] (*Sr8*)	15B	69	139	36	4:9:3	.20 — .30
	56	97	24	10	48:13:3	.20 — .30
X Kenya 117A — Mar.[6] (*Sr9*)	56	73		6	61:3	.20 — .30
X Egypt Na95 — Mar.[4] (*Sr10*)	56	150		15	55:9	.05 — .10
X Rio Negro (*Sr8, Sr9*)	56	63		0		
Kenya 341						
X Kenya 58 — Mar.[6] (*Sr6*)	15B	113		0		
	56	171		0		
X Red Egyptian — Mar.[6] (*Sr8*)	15	35	84	30	4:9:3	.50 — .95
	56	182	50	22	48:13:3	.01 — .02[1]
X Kenya 117A — Mar.[6] (*Sr9*)	56	81		1	61:3	.10 — .20
X Egypt Na95 — Mar.[4] (*Sr10*)	56	159		22	55:9	.30 — .50

[1]The poor fit is probably the result of a few plants heterozygous for *Sr8* being classified as susceptible

TABLE 5. — RESULTS OF SEEDLING TESTS ON F_2 FAMILIES FROM THE BACKCROSSES OF MAGNIF G, FRONTANA AND RIO NEGRO TO MARQUIS

		Race 15B Number of families		Totals (race 56)	P (1:1:1:1)	Ratios within segregating families
		Seg. 3MR:1S	S			
Magnif G-Marquis² F_2						
	Seg. 3MR:1S	9	0	9		167MR:63S – P(3:1) = .30-.50
	Seg. 15MR:1S	16	0	16		446MR:27S – P(15:1) = .30-.50
Race	Seg. 3MR:1S	0	12	12	.30-.50	194MR:55S – P(3:1) = .20-.30
56	Susc.	0	8	8		
	Totals (race 15B)	25*	20	45		*541MR:186S – P(3:1) = .50-.95
	P(1:1)	.50-.70				
Frontana-Marquis² F_2						
	Seg. 3MR:1S	12	0	12		250MR:81S – P(3:1) = .50-.95
	Seg. 15MR:1S	13	0	13	.90-.95	387MR:26S – P(15:1) = 1.0
Race	Seg. 3MR:1S	0	14	14		268R:70S – P(3:1) = .05-.10
56	Susc.	0	11	11		
	Totals (race 15B)	25**	25	50		**525MR:170S – P(3:1) = .50-.95
	P(1:1)	1.0				
Rio Negro-Marquis² F_2						
	Seg. 3MR:1S	12	0	12		230MR:76S – P(3:1) = 1.0
	Seg. 15MR:1S	13	0	13		318MR:29S – P(15:1) = .10-.20
Race	Seg. 3MR:1S	0	18	18	.10-.20	247MR:106S – P(3:1) = .02-.05 (the poor fit is largely due to 2 of the 18 families)
56	Susc.	0	8	8		
	Totals (race 15B)	25***	26	51		***518MR:194S – P(3:1) = .10-.20
	P(1:1)	1.0				

This gene is relatively weak, conditioning a 2-2$^+$ type reaction, and is nearly recessive. Segregations, however, were often not clear-cut. In tests where the reactions seemed to be well defined reasonable fits to a 1:3 ratio were obtained. Under other conditions, too many plants gave intermediate reactions and it was evident that some of the heterozygotes were moderately resistant. Some families in this group were tested as often as five times.

The results of test crosses to identify the genes carried by the three varieties are given in Table 4. The data show clearly that the gene conditioning a high degree of resistance is *Sr6*. The second gene behaves like *Sr10*; however the F_2 populations from crosses between the three varieties and a line supposedly carrying *Sr10* all segregated. Unfortunately there is some doubt [see Green *et al.* (2)] as to whether this lines carries *Sr10*. Although *Sr10* was originally identified using race 56, the Marquis backcross line gives very erratic results with this race. Consequently, the results cannot be considered conclusive.

The gene *Sr6* behaved somewhat differently in the crosses involving the three varieties than in material studied previously. Always before it had been completely dominant with race 56 but completely recessive with race 15B. In this material, however, plants that were homozygous gave a fleck reaction to both races but the heterozygotes gave type 1 to type 3$^-$

TABLE 6. — RESULTS OF SEEDLING RUST TESTS ON F_2 POPULATIONS FROM TEST CROSSES INVOLVING MAGNIF G, FRONTANA AND RIO NEGRO

Cross	Race	Number of plants		Expected ratio	P
		MR	S		
Magnif G					
X Red Egyptian — Mar.[6] (*Sr8*)	15B	161	0		
	56	161	0		
X Kenya 117A — Mar.[6] (*Sr9*)	56	241	0		
X Egypt Na95 — Mar.[4] (*Sr10*)	56	72	1	61:3	.10 — .20
X Egypt Na95 (*Sr7*, *Sr9*, *Sr10*)	56	160	0		
Frontana					
X Magnif G	15B	75	0		
	56	77	0		
X Red Egyptian — Mar.[6] (*Sr8*)	15B	219	0		
	56	202	0		
X Kenya 117A — Mar.[6] (*Sr9*)	56	221	0		
X Egypt Na95 — Mar.[4] (*Sr10*)	56	123	7	61:3	.50 — .95
X Egypt Na95 (*Sr7*, *Sr9*, *Sr10*)	56	163	0		
Rio Negro					
X Magnif G	15B	126	0		
	56	121	0		
X Frontana	15B	124	0		
	56	119	0		
X Kenya 117A — Mar.[6] (*Sr9*)	56	112	0		
X Egypt Na95 — Mar.[4] (*Sr10*)	56	274	6	61:3	.05 — .10
X Egypt Na95 (*Sr7*, *Sr9*, *Sr10*)	15B	137	7	15:1	.30 — .50
	56	145	0		

reactions with race 56. This made it impossible to determine with certainty which of the families that segregated for *Sr6* also segregated for the recessive gene giving moderate resistance to race 56.

Two of the varieties, Kenya N.B. 263 and Kenya 341, also segregated for a third gene conditioning a type 2$^+$ reaction only to race 15B. The segregations were not clear in the backcrosses to Marquis and are not recorded in the tables. Often it appeared that the first seedling leaf was considerably more resistant than the second. Several F_3 families were grown and clearly resistant plants were recovered. The gene is being transferred to Marquis.

Magnif G, Frontana and Rio Negro

Magnif G, Frontana and Rio Negro behaved alike and will be considered together. The data from the backcrosses to Marquis are given in Table 5. In tests with race 15B, half the families segregated for a moderately resistant (type 2) reaction and half were susceptible. Furthermore, each family that segregated with race 15B also segregated for a type 2 reaction to race 56. Clearly one dominant gene is conditioning resistance to both races. Of those families that were susceptible to race 15B about half segregated for a moderately resistant (type 2) reaction to race 56. All three varieties, therefore, carry a second gene that conditions resistance only to race 56.

The results of test crosses used to identify the genes carried by Magnif G, Frontana and Rio Negro are given in Table 6. When any one of the three was crossed with a parent carrying either *Sr8* or *Sr9*, all of the F_2 progeny were moderately resistant to race 56. All three varieties, therefore, carry *Sr8* and *Sr9*. As expected, all of the F_2 progeny of crosses between Frontana, Magnif G and Rio Negro were moderately resistant to both races.

Sapporo

Sapporo has fairly good resistance to race 15B but is moderately susceptible to race 56. The results of tests with race 15B on the families from backcrosses to Marquis are given in Table 7. Approximately half the

TABLE 7. — RESULTS OF SEEDLING TESTS WITH RACE 15B ON F_2 FAMILIES FROM THE BACKCROSS OF SAPPORO TO MARQUIS

	Segregating 3 resist. : 1 susc.	Susceptible
Number of Families	29[1]	17
Expected (1:1)	23	23
P	.05 — .10	

[1]The segregation within the families was:
732R:248S plants P for a 3:1 ratio = .50-.95

families segregated although the probability of a fit to a 1:1 ratio is somewhat low. Within segregating families an excellent fit to a ratio of 3 resistant: 1 susceptible plant was obtained. It appears, therefore, that Sapporo carries one dominant gene for resistance to race 15B. As a further check additional backcrosses were made and the F_1 plants tested with race 15B. Thirty-one plants were resistant and twenty-five susceptible, a good fit to the expected 1:1 ratio.

The Sapporo resistance to race 15B looked very much like that conditioned by *Sr7*. A cross was made between Sapporo and Kenya Governor (*Sr7*) and 204 F_2 plants proved to be resistant to race 15B. Sapporo, therefore, carries *Sr7*.

Kentana 52

Kentana has a high degree of resistance to races 15B and 56. The data from tests on the backcrosses to Marquis are given in Table 8. Approximately half the families segregated for a very resistant (fleck) reaction to

TABLE 8. — RESULTS OF SEEDLING RUST TESTS ON F_2 FAMILIES FROM THE BACKCROSS OF KENTANA 52 TO MARQUIS

	Race 15B Number of families				Totals (race 56)	Expected (1:1)	P
	Seg. 1VR:3S	Seg. 13VR-MR:3S	Seg. 3MR:1S	Susc.			
Race 56 Seg. 3VR:1S	16	14	0	0	30[1]	24.5	.10 — .20
Susc.	0	0	10	9	19	24.5	
Totals (race 15B)	16[2]	14[3]	10[4]	9	49		
Expected (1:1:1:1)	12.25	12.25	12.25	12.25			
P	.30 — .50						

The segregations within families were as follows:
Race 56
[1]766VR:252S P for a 3:1 ratio = .50-.95
Race 15B
[2]78VR:238S P for a 1:3 ratio = .50-.95
[3]178VR-MR:38S P for a 13:3 ratio = .50-.95
[4]227MR:69S P for a 3:1 ratio = .50-.95

TABLE 9. — RESULTS OF SEEDLING RUST TESTS ON F_2 POPULATIONS FROM TEST CROSSES INVOLVING KENTANA 52

Cross	Race	Number of plants			Expected ratio	P
		VR	MR	S		
Kentana 52						
X Kenya 58 — Mar.[6] (*Sr6*)	15B	269		0		
X Kenya Governor (*Sr7*)	15B	210		0		
X Frontana	15B	156		3	61:3	.05 — .10
	56	156		2	63:1	1.0

TABLE 10. — RESULTS OF SEEDLING RUST TESTS ON F^2 FAMILIES FROM THE BACKCROSS OF NO. 466 TO MARQUIS

		Race 15B Number of families		Totals (race 56)	Expected (1:2:1:2:1:1)	P
		Seg. 1VR:3S	Susc.			
Race 56	Seg. 3VR:1S	6	0	6[1]	6.6	.50 — .95
	Seg. 12VR:3MR:1S or 48VR:13MR:3S	13	0	13[2]	13.3	
	Seg. 12VR:1MR:3S	8	0	8[3]	6.6	
	Seg. 3MR:1S or 13MR:3S	0	16	16[4]	13.3	
	Seg. 1MR:3S	0	5	5[5]	6.6	
	Susc.	0	5	5	6.6	
Totals (race 15B)		27	26[6]	53	53.0	
Expected (1:1)		26.5	26.5			
P		1.0				

The ratios within segregating families were as follows:
Race 56
[1]260VR:93S plants — P for a 3:1 ratio = .50-.95
[2]629VR:144MR:41S plants — P for a 48:12.5:3.5 ratio = .30-.50 (assuming half 12:3:1 and half 48:13:3)
[3]449VR:46MR:117S plants — the separation of moderately resistant and susceptible plants was not sufficiently clear to be reliable.
[4]750MR:194S plants — P for a 12.5:3.5 ratio = .30-.50 (assuming half 3:1 and half 13:3)
[5]The separation was not sufficiently clear to be reliable.
Race 15B
[6]423VR:1207S plants — P for a 1:3 ratio = .30-.50

both races. The fleck reaction was the only type of resistance obtained with race 56. With race 15B, however, half the families also segregated for a moderately resistant reaction (type $1^+ - 3^-$). Evidently one gene conditions high resistance to both races, while a second controls moderate resistance to race 15B only. The segregations within families show that the first gene is dominant with race 56 but recessive with race 15. The second gene is partially dominant.

Since Kenya C9906, which is one of the parents of Kentana 52, carries genes *Sr6* and *Sr7* it seemed likely that Kentana 52 carries one or both. The results of test crosses with lines carrying *Sr6* and *Sr7* are given in Table 9. The data show clearly that the two genes in Kentana 52 are *Sr6* and *Sr7*. Since Frontana is one of the parents of Kentana 52, it appeared that Frontana and Kentana 52 could have genes in common. However, the F_2 plants from the cross between them segregated with both races.

No. 466

The results of tests on the F_2 families from backcrosses of No. 466 to Marquis are given in Table 10. With race 15B half the families segregated for a very resistant (fleck) reaction and each family also segregated for a

fleck reaction to race 56. Evidently one gene conditions resistance to both races. The segregations within families show that it is dominant with race 56 but recessive with race 15B. Although it is not shown in Table 10, 24 of the families had some plants with a 2-2+ reaction to race 15B. Most of the families were tested two to four times and the results were not entirely consistent. The F_2 of a cross with Marquis segregated 14VR:14MR:38S, a satisfactory fit to a 4:3:9 ratio. It appears that a recessive gene giving a very moderate type of resistance is present. The gene is now being transferred to Marquis.

The results with race 56 are much more complex. As already mentioned half the families segregated for a fleck reaction. Of the families that were susceptible to race 15B, three-quarters (21/26) segregated for moderate resistance to race 56. Within families, ratios of 3:1, 1:3 and 13:3 were recorded. In some cases the numbers were too small to make it possible to distinguish definitely between a 3:1 and a 13:3 ratio. These two classes have, therefore, been grouped in the table. It seems clear that each of two genes conditions moderate resistance to race 56. One is dominant and the other nearly recessive. However, segregations involving the recessive gene were not sufficiently clear to be reliable. Where both genes segregated in the same family the reactions seemed to be more distinct and the results have been given in the table.

The results of test crosses to identify the genes carried by No. 466 are given in Table 11. The data show clearly that the variety carries *Sr6*. One of the genes in No. 466 behaves exactly like *Sr9*. However, in the test cross with *Sr9*, one of the 309 plants was susceptible. If No. 466 does not carry *Sr9* then the cross should segregate for 4 genes, 3 dominant and 1 recessive, and give a ratio of 253:3. The actual segregation will fit this ratio. Additional data were obtained from a cross with Rio Negro which carries *Sr9* as well as *Sr8*. With race 56, 169 F_2 plants were tested and gave reactions ranging from 0;—1+. The high degree of resistance suggests that the varieties have a gene in common. It is concluded, therefore, that No. 466 carries *Sr9* and that the one susceptible plant in the test cross came from a stray seed. Almost the same thing happened in the test cross with *Sr10* where one of 365 plants was susceptible. In this case,

TABLE 11. — RESULTS OF SEEDLING RUST TESTS ON F_2 POPULATIONS FROM TEST CROSSES INVOLVING NO. 466

Cross	Race	Number of plants			Expected ratio	P
		VR	MR	S		
No. 466						
X Kenya 58-Mar.[6] (*Sr6*)	15B	210		0		
X Kenya 58-That.[10] (*Sr6*)	15B	103		0		
X Red Egyptian-Mar.[6] (*Sr8*)	15B	35	65	18	4:9:3	.30-.50
X Kenya 117A-Mar.[6] (*Sr9*)	56	308		1*	309:0	
X Egypt Na95-Mar.[4] (*Sr10*)	56	364		1*	365:0	

*See text

however, if No. 466 does not carry *Sr10*, then the cross should segregate for 2 dominant and 2 recessive genes, and give a ratio of 247:9. The results clearly do not fit this ratio and it is concluded that the susceptible plant was a stray. The third gene in No. 466 must be *Sr10*.

P.I.60599

P.I.60599 normally gives a 0;—1⁻ reaction to races 15B and 56. It proved to be the most difficult of the varieties to analyse for two reasons. First, it carried more genes than any other variety and very complex ratios were obtained. Second, it gave partially sterile hybrids in many crosses, particularly those with Egypt Na95, Kenya Governor and Sapporo. Cytological studies were made on F_1 plants from crosses between P.I.60599 and these three varieties. In the hybrids with Egypt Na95 and Kenya Governor the normal metaphase configuration was $18_{II} + 1_{VI}$. The hybrids with Sapporo frequently had cells with metaphase configurations of either $15_{II} + 1_{VIII} + 1_{IV}$ or $15_{II} + 3_{IV}$. Apparently, P.I.60599 differs from the other varieties by two or more translocations. The partial sterility of hybrids could result from meiotic irregularity.

Some sterility occurred in hybrids with Marquis but adequate seed was obtained in backcrosses. The results of tests on the backcrosses are given in Table 12. In the tests with race 15B only 4 of 47 backcross families were susceptible. The results will fit both a 7:1 and a 15:1 ratio suggesting that either 3 or 4 genes are segregating. With race 56 the segregation was also 43 segregating:4 susceptible families but different families were susceptible. The segregations within families suggested that for each race only three genes were involved. Furthermore, it also appeared that one gene conditioned a type 2 reaction to both races. This means, then, that P.I.60599 carries five genes, two giving resistance only to race 15B, two giving resistance only to race 56 and one giving resistance to both races. All of the genes conditioned intermediate types of reactions and with each race resistant reactions ranged from 1-2⁺. For this reason it was impossible to sort the segregating families into groups.

TABLE 12. — RESULTS OF SEEDLING TESTS ON F_2 FAMILIES FROM THE BACKCROSS OF P.I.60599 TO MARQUIS

	Race 15B Number of families		Totals (race 56)	Expected (7:1)	P
	Seg.	S			
Race 56 Seg.	39	4	43	41.1	.30–.50
Race 56 S	4	0	4	5.9	
Totals (race 15B)	43	4	47	47	
Expected (7:1)	41.1	5.9	47		
P	.30–.50				

TABLE 13. — THE RESULTS OF SEEDLING RUST TESTS ON F_2 POPULATIONS FROM TEST CROSSES INVOLVING P.I.60599

Cross	Race	Number of plants		Expected ratio	P
		R	S		
P.I. 60599					
X Kenya 58-Mar.[6] (*Sr6*)	15B	151	4	253:3	.10
	56	159	2	253:3	1.00
X Egypt Na101-Mar.[6] (*Sr7*)	15B	37	0		
X Egypt Na95 (*Sr7, Sr9, Sr10*)	15B	60	0		
	56	63	0		
X Egypt Na101 (*Sr7*)	15B	19	0		
X Sapporo (*Sr7*)	15B	40	0		
X Red Egyptian-Mar.[6] (*Sr8*)	15B	307	0		
	56	317	0		
X Kenya 117A-Mar.[6] (*Sr9*)	56	313	0		
	15B	163	2	63:1	.50-.95
X Egypt Na95-Mar.[4] (*Sr10*)	56	454	0		

The results of test crosses to identify the genes carried by P.I.60599 are given in Table 13. Neither P.I.60599 nor any of the backcross families gave the hypersensitive reaction typical of *Sr6* and the test cross shows that the variety does not carry this gene. Twenty-six of the backcross families contained plants giving the reaction typical of *Sr7* — 1-3⁻ type pustules with considerable yellowing. Unfortunately, some of the test crosses with lines carrying *Sr7* were highly sterile. However, 156 plants from 4 crosses were tested and all gave a reaction typical of *Sr7*. There seems to be little doubt that P.I.60599 carries *Sr7*. The test crosses also show that the gene conditioning a 2 reaction to both races is *Sr8* and that the two genes conditioning resistance only to race 56 are *Sr9* and *Sr10*. The one remaining gene conditions a type 2 reaction to race 15B and is different than any of the previously named genes.

DISCUSSION

The 11 common wheat varieties of widely diverse origin were studied in an attempt to locate new genes for rust resistance. In this respect the work largely failed. The major genes for resistance carried by the 11 varieties had all been identified previously. The probable genotype of each variety is given in Table 14. Four varieties, Kenya N.B. 263, Kenya 341, No. 466 and P.I.60599, contain a gene which had not previously been identified and which conditions moderate resistance to race 15B. Since it is not known whether the genes are the same or different in each variety, they have not yet been assigned symbols. Three of the Kenya varieties, Kenya 122, Kenya N.B. 263 and Kenya 341, carry a gene which conditions moderate resistance to race 56 and behaves much like *Sr10*. The genes from both groups of varieties are being isolated in Marquis and when this has been done the appropriate lines will be intercrossed to test for allelism.

The results obtained in this study tend to strengthen the conclusion that the number of genes controlling stem rust resistance in common wheat

TABLE 14.—PROBABLE GENOTYPES FOR THE 11 VARIETIES

Variety	Genotype
Kenya 318	*Sr6 Sr6*
Kenya 122[1]	*Sr6 Sr6*
Kenya N.B.263[1]	*Sr6 Sr6*
Kenya 341[1]	*Sr6 Sr6*
Kentana 52	*Sr6 Sr6* *Sr7 Sr7*
Magnif G	*Sr8 Sr8* *Sr9 Sr9*
Frontana	*Sr8 Sr8* *Sr9 Sr9*
Rio Negro	*Sr8 Sr8* *Sr9 Sr9*
Sapporo	*Sr7 Sr7*
No. 466[1]	*Sr6 Sr6* *Sr9 Sr9* *Sr10 Sr10*
P.I.60599[1]	*Sr7 Sr7* *Sr8 Sr8* *Sr9 Sr9* *Sr10 Sr10*

[1]Kenya 122, Kenya N.B.263, and Kenya 341 each carry an additional gene which conditions moderate resistance to race 56. Kenya N.B.263, Kenya 341, No. 466 and P.I.60599 each carry an additional gene giving moderate resistance to race 15B (*see text*).

is relatively small. The new gene or genes that were discovered are of doubtful importance. In order to build up a supply of genes for rust resistance it will probably be necessary to make much greater use of related species and genera. Present wheat breeding for resistance to race 15B appears to be based largely on three genes, *Sr6*, *Sr7* and *Sr8*, except where genes are being transferred from other species.

In general, the results in this paper agree with those reported for the same varieties by other authors. However, only a few of the varieties had been studied previously.

Goulden and Stevenson (1) reported that Kenya 122 (R.L. 1373) has one gene giving resistance to race 15B. This gene has now been identified as *Sr6*. Pugsley (6) showed that Kenya 122 and Kenya N.B. 263 have a gene, *SrKa1* in common and the present work shows that both carry *Sr6*. Green *et al.* (2) have suggested that *SrKa1* and *Sr6* are the same.

Wiggin (7) analysed Kentana 52, using the Chinese spring monosomics. He concluded that it carries two dominant, linked genes on chromosome XX, one giving resistance to race 56, the other to race 15B. The present study indicates that Kentana 52 carries *Sr6* which is on chromosome XX and *Sr7* which is on chromosome VIII. Since *Sr6* is dominant with race 56 but recessive with race 15B, heterozygotes will be susceptible to 15B but resistant to race 56. Plants which carry *Sr7* but not *Sr6* will be resistant to race 15B but susceptible to race 56. Thus, the apparent occurrence of two cross-over classes can be accounted for and Wiggin's results reconciled with those reported here.

Burnham* worked with Frontana and found that it had one gene on chromosome VI. With one culture of race 56 the presence of a second gene was indicated. The results with race 56 reported in this paper show that Frontana carries two genes, *Sr8* (on chromosome VI) and *Sr9*.

An important feature of the present work is the use of test crosses involving Marquis lines carrying single genes for rust resistance. Marquis

*Private communication

backcrosses, carrying genes *Sr6*, *Sr7*, *Sr8*, *Sr9*, *Sr10* and *Sr11*, are now available. Their use makes it relatively easy to determine whether a variety carries any of the known genes for resistance. All of the lines except *Sr10* give reasonably clear reactions to various races [see Green *et al.* (2)]. It is not certain that the *Sr10* line actually carries *Sr10* since it gives rather erratic results with race 56, although this is the race with which it was originally identified. The line does, however, have more resistance than Marquis. A new attempt to isolate *Sr10* is under way and after two backcrosses to Marquis, clear resistance (type 2) has been maintained.

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COMPARISON OF GRASSES FOR DRYLAND TURF[1]

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ABSTRACT

Russian wild-rye, Fairway crested wheatgrass, and certain strains of sheep fescue showed superior hardiness, resistance to wear, and turf quality at Saskatoon, Sask., Streambank wheatgrass, western wheatgrass, and a native sedge were persistent but showed inferior turf quality. Grasses lacking hardiness included Merion bluegrass, Kentucky bluegrass, Canada bluegrass, creeping red fescue, chewing's fescue, and certain strains of sheep fescue. Bromegrass had unsatisfactory turf quality and was easily damaged by vehicular traffic.

INTRODUCTION

Hardy, drought-tolerant turf grasses are needed for the open plains area of Western Canada where supplementary water is not available. Potential uses for turf include school-yards, playing-fields, cemeteries, roadsides, landing-strips, machinery yards, and irrigation ditch banks. Quality of turf often is not a major consideration, providing a cover is obtained which needs little maintenance.

The suitability of the Fairway variety of crested wheatgrass, *Agropyron cristatum* (L.) Gaertn., for dryland turf was noted by Kirk in 1932 (3). Russian wild-rye, *Elymus junceus* Fisch., recently was found to be superior to Fairway crested wheatgrass for turf, especially in its ability to remain green throughout the summer (2). Sheep fescue, *Festuca ovina* L., was recommended for dryland turf in Washington because of its durability and drought tolerance (4). Streambank wheatgrass, *Agropyron riparium* Scribn. and Smith, showed qualities suggesting its use for dryland turf and irrigation ditch banks (1). These grasses and others of potential value for turf were tested comparatively at Saskatoon from 1951 to 1960 for hardiness, turf quality, and durability.

MATERIALS AND METHODS

Roadways of an experimental plot area were used to test grasses under traffic conditions. Plots were arranged in a linear fashion along roads so that they received similar amounts of vehicle traffic. Four tests were established from 1951 to 1956 and were observed for the period 1951 to 1960. Size of plots varied from 8 x 20 feet to 40 x 20 feet and plots were arranged in duplicate randomized blocks in each test. Seeding was done in May and June on fallow land. Seed was broadcast and raked in by hand, and the land packed with a field packer. No watering or fertilization was provided. Plots were mowed periodically throughout the summer with a sickle-type field mower to control weeds and growth of grasses.

Seed for species in commercial use was obtained from local seed houses. Streambank wheatgrass was represented by the Sodar variety. The S-3482 strain of sheep fescue was obtained from the Soil Conservation Service, Pullman, Washington. This strain was classed as "hard fescue", *Festuca ovina, duriuscula* (L.) Koch. The S-885 strain of sheep fescue was obtained through the Central Experimental Farm, Ottawa, Ont., from an

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uncertain source. Other strains of sheep fescue traced to introductions from Kazakstan, U.S.S.R. Of these the S-1748 strain appeared similar to the hard fescue strain S-3482 from Washington.

Involute-leaved sedge, *Carex eleocharis* Bail, and western wheatgrass, *Agropyron smithii* Rydb., were transplanted as clones from native prairie in the vicinity of Saskatoon.

In scoring grasses for turf quality, consideration was given to density of turf, freedom from weeds, and colour at various times throughout the summer. Resistance to wear was noted in areas of plots where wheel traffic was concentrated and ratings reflect the ability of grasses to maintain ground cover.

RESULTS

Comparison of Grasses Seeded 1951

Grasses were seeded at a rate of 1 pound per 1,000 square feet. This same rate of seeding was used for mixtures with equal proportions of each grass in the mixture. Stands were complete for all species, except Kentucky bluegrass which showed only 70 per cent cover. Severe winter killing occurred in the winter of 1953-1954. Turf quality was scored in 1956 and final stand records were made in 1959. As traffic over this test was light no durability ratings were made. Results of this test are summarized in Table 1.

TABLE 1.—COMPARISON OF GRASSES FOR TURF. SEEDED 1951.

Grass and symbol	Winter kill 1954	Turf quality 1956	Stand 1959
	%		%
Kentucky blue (KB)	17	Good	28
Creeping red fescue (Cr)	45	Poor	0
Chewing's fescue (Ch)	77	Poor	0
Sheep fescue S-885 (Sh)	15	Good	58
Brome (Br)	5	Poor	30
Fairway crested wheat (Cwg)	0	Fair	100
Mixture — KB and Cr	27	Good	25
Mixture — KB and Br	5	Good	55
Mixture — KB and Cwg	0	Fair	100
Mixture — Cr and Br	5	Poor	25
Mixture — Cr and Cwg	0	Fair	100
Mixture — KB, Cr, Br, Cwg	0	Fair	100

Kentucky bluegrass and sheep fescue showed best turf quality but lacked hardiness. Crested wheatgrass was fully hardy and maintained good stands until 1960, but quality of turf was mediocre, especially during dry summers. Chewing's fescue was severely winter killed in 1954. Creeping red fescue showed serious winter damage in 1954, and was eliminated by 1956. Bromegrass was hardy but the open nature of the turf resulted in an unattractive appearance. Mixtures tended to be dominated by one component. Grasses in the order of aggressiveness in mixtures were Fairway crested wheatgrass, Kentucky bluegrass, bromegrass, and creeping red fescue.

TABLE 2.—COMPARISON OF TURF GRASSES FOR PERSISTENCE, WEARING ABILITY, AND TURF QUALITY. SEEDED 1953.

Grass and symbol	Original stand	Winter kill 1954	Wearing ability	Turf rating 1956	Stand 1959
	%	%			%
Kentucky blue (KB)	55	7	Good	Good	15
Merion blue	42	35	Good	Fair	0
Canada blue	15	35	Fair	Fair	0
Creeping red fescue (Cr)	95	65	Poor	Fair	0
Chewing's fescue	75	95	—	—	0
Streambank wheat	100	0	Good	Fair	68
Russian wild-rye	100	0	Excellent	Fair	100
Brome (Br)	100	0	Poor	Poor	35
Fairway crested wheat (Cwg)	100	0	Excellent	Fair	100
Involute-leaved sedge	15	0	Excellent	Poor	95
Sheep fescue S-885 (Sh)	90	2	Fair	Good	15
Sheep fescue S-1722	70	0	Good	Excellent	70
Sheep fescue S-1733	80	0	Good	Excellent	70
Sheep fescue S-1748	100	0	Good	Excellent	80
Sheep fescue S-1758	75	0	Excellent	Good	100
Sheep fescue S-1765	85	0	Good	Excellent	90
Sheep fescue S-1792	85	0	Excellent	Excellent	100
Mixture — KB and Br	95	0	Fair	Poor	10
Mixture — KB and Cr	85	20	Fair	Fair	5
Mixture — Cr and Sh	45	0	Fair	Fair	15
Mixture — Cr and Br	95	0	Poor	Poor	10

Comparison of Grasses Seeded 1953

Grasses were seeded at a rate of 3 pounds per 1,000 square feet and this resulted in good stands except for the three bluegrasses. Involute-leaved sedge was planted as cuttings in rows 1 foot apart. Fairly complete cover was obtained for this species by 1958. Results for this test appear in Table 2.

Severe winter killing was noted in the spring of 1954 for creeping red fescue and chewing's fescue. Merion and Canada bluegrass also showed considerable damage, being inferior to Kentucky bluegrass in hardiness. Reduced stands in 1959 reflect winter damage in subsequent years combined with drought and traffic damage. Outstanding persistence and durability was shown by Russian wild-rye. Fairway crested wheatgrass and certain strains of sheep fescue maintained complete stands but showed more damage from traffic than Russian wild-rye. Streambank wheatgrass and involute-leaved sedge were quite durable under traffic, but neither grass presented much turf.

Comparison of Grasses Seeded 1954

Seeding rates were not recorded for this test. The location was such that little traffic was received. However, winter killing and drought caused severe reduction of stands. Russian wild-rye and Fairway crested wheatgrass were markedly superior to the other grasses.

Comparison of Grasses Seeded 1956

A seeding rate of 2.5 pounds per 1,000 square feet was used for all grasses in this test. Good stands were obtained except for Merion blue-

TABLE 3.—COMPARISON OF TURF GRASSES FOR SURVIVAL AND TURF QUALITY. SEEDED 1954.

Grass	Stand 1956	Winter kill 1957	Turf rating 1958	Stand 1959
	%	%		%
Kentucky blue	80	5	Fair	8
Merion blue	78	20	Fair	10
Canada blue	75	7	Fair	10
Fairway crested wheat	90	0	Good	100
Russian wild-rye	70	0	Good	100
Sheep fescue, S-885	65	7	Fair	5
Brome	70	0	Poor	15

TABLE 4.—COMPARISON OF TURF GRASSES FOR SURVIVAL, PERSISTENCE, AND QUALITY. SEEDED 1956.

Grass and symbol	Stand 1956	Winter kill 1959	Wearing ability	Turf rating 1959
	%	%		
Kentucky blue (KB)	95	20	Fair	Fair
Merion blue (MB)	30	20	Fair	Poor
Delta blue	85	10	Fair	Fair
Creeping red fescue (Cr)	72	50	Fair	Poor
Fairway crested wheat	100	0	Excellent	Good
Russian wild-rye	100	0	Excellent	Good
Western wheatgrass	100	0	Good	Fair
Sheep fescue S-885 (Sh)	87	20	Fair	Poor
Sheep fescue S-1722	70	0	Good	Good
Sheep fescue S-1733	95	0	Fair	Good
Sheep fescue S-1748	88	5	Good	Good
Sheep fescue S-1758	77	10	Good	Fair
Sheep fescue S-1765	90	5	Good	Good
Sheep fescue S-3482	92	5	Good	Good
Mixture, Cr and KB	82	20	Fair	Fair
Mixture, Cr and MB	85	75	Fair	Poor
Mixture, Sh and KB	75	40	Fair	Poor

grass. Sod from native prairie containing a high proportion of western wheatgrass was used instead of seed for the establishment of this species. Traffic was fairly heavy and considerable thinning in the centre portions of plots occurred by 1959 (Table 4).

The bluegrass strains, creeping red fescue, and some strains of sheep fescue showed winter killing. There was an indication that the hard fescue strains S-1748 and S-3482 were not as hardy as certain strains of true sheep fescue such as S-1722 and S-1733. Russian wild-rye and Fairway crested wheatgrass showed excellent wearing ability. Some of the fescue strains showed good wearing ability and somewhat better turf quality than Fairway crested wheatgrass and Russian wild-rye.

DISCUSSION

Russian wild-rye and Fairway crested wheatgrass showed great tolerance to drought and frost, and withstood heavy traffic. They established quickly and gave good weed control after the year of seeding without

the use of herbicides. The leaves of these grasses are coarse in comparison with the bluegrasses and fescues, and the leaves of Russian wild-rye are glaucous in colour. Russian wild-rye showed slightly better tolerance of traffic than Fairway crested wheatgrass and remained greener in the late summer and fall. Rates of seeding of 2 pounds per 1,000 square feet for Fairway crested wheatgrass and 3 pounds per 1,000 square feet for Russian wild-rye gave satisfactory stands. These species can be recommended for turf in the Brown and Dark Brown Soil Zones.

Certain strains of sheep fescue approached crested wheatgrass and Russian wild-rye in hardiness and resistance to wear. Sheep fescue provided a better quality of turf than crested wheatgrass and Russian wild-rye. Growth of sheep fescue was short and when seeded heavily there was little tendency to head out. Consequently, mowing was seldom required. Most strains of sheep fescue were bluish or glaucous in colour which made them less attractive for turf, especially in mixtures. The so-called "hard" fescue strains S-3482 and S-1748 were bright green in colour, similar to Kentucky blue or creeping red fescue. A seeding rate of 3 pounds per 1,000 square feet appeared necessary to ensure complete stands of these fescues. Growth of seedlings was slower than that of crested wheatgrass, and weed control was a problem in the first and second years of establishment. Strains of sheep fescue, such as S-1758 or S-1792, and strains of hard fescue, such as S-1748 or S-3482, appear worthy of multiplication for commercial use in Western Canada.

Involute-leaved sedge was hardy and persistent under traffic in these tests, but it provided little turf. Since seed of this plant is difficult to harvest it is improbable that it will be used on a commercial scale. However, where this species is present in substantial amounts in native prairie it might be preserved as turf. The Sodar strain of streambank wheatgrass and native western wheatgrass were only moderately satisfactory for turf in these tests. Quality of turf provided by these grasses was inferior to that of Fairway crested wheatgrass, Russian wild-rye, and sheep fescue.

Severe winters and drought limit the use of better quality turf grasses such as Kentucky bluegrass and creeping red fescue for unwatered prairie areas. Chewing's fescue was especially susceptible to winter damage in these tests. Merion bluegrass and Canada bluegrass appeared less hardy than Kentucky bluegrass under these conditions of limited moisture.

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INHERITANCE OF QUALITY AS RELATED TO AGRONOMIC CHARACTERS IN ADVANCED LINES OF A SPRING WHEAT CROSS[1]

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ABSTRACT

The expansion, sedimentation and mixograph flour quality tests were used to evaluate the bread-making properties in F_8 and F_9 spring wheat lines, derived from a cross between the poor quality hybrid R.L.2265.46 (a McMurachy x Exchange selection) and the variety Redman. The results from these quality tests were considered in relation to head type and stem rust resistance to race 15B-4 (Can.) which is controlled by gene *Sr6* present in the numbered parent.

In general, the lines with compact spike and stem rust resistance showed inferior quality when compared with those having fusiform heads and with those which were susceptible to rust. It was observed that none of the lines of one population had good quality whereas in the second population, where the R.L. 2265-46 parent was shown to be heterozygous for both head type and rust resistance, 2.0 per cent of the population was promising in quality, rust resistance and yield. The *Sr6* gene was found to be loosely linked with the gene for compact spike. The mean yields of lines having compact spikes were significantly lower than those with fusiform spikes.

INTRODUCTION

In the breeding of Selkirk hard red spring wheat, a stem rust-resistant variety R.L.2265 (McMurachy x Exchange) was used as the parent contributing the gene *Sr6* (7) for resistance to stem rust race 15B and to some of its biotypes to which Redman, the other parent, was susceptible.

For this study a similar cross to that used in the production of Selkirk was made using R.L.2265.46, a selection of poor quality, low in yield and with compact type head, and Redman, a high quality, high yielding bread wheat with a tapering fusiform head. As difficulties were found in obtaining a suitable line with good quality and yield from a single cross, in breeding the variety Selkirk, the backcross system using Redman three times as the recurrent parent was followed. Through the repetition of this similar cross and the study of the progeny with respect to quality and agronomic characteristics it is hoped to ascertain some of the reasons for these difficulties. Three different tests for flour quality were used, viz., expansion, mixograph and sedimentation. The data from these tests were related to the agronomic characters – resistance to stem rust 15B-4 (Can.), head type, and grain yield.

Data from the expansion test (8, 9) and from the sedimentation test (11) have given high correlations with loaf volume data and are considered to give a good expression of the potential baking strength of flour. The mixograph test is a physical dough test (4, 5) giving a pattern in a mixogram which denotes the mixing characters of a flour-water dough. Strong flours exhibit considerable resistance to mixing and weaker types lose this resistance early in the mixing.

Race 15B-4 (Can.)[2] stem rust to which R.L.2265-46 is resistant and Redman is susceptible in the seedling stage, was used to test the reaction of the progeny lines.

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[1]Contribution No. 49, Genetics and Plant Breeding Research Institute, Canada Department of Agriculture, Ottawa, Ont.

[2]Whenever stem rust is mentioned, reference is made only to race 15B-4 (Can.).

MATERIAL AND METHODS

The cross between R.L.2265-46 and Redman was first made in the field in 1949, and the F_1 and F_2 were grown in the greenhouse in 1949 and in 1950. Many F_2 plants failed to set seed during the winter as a result of low light intensity in the greenhouse. However, 240 plants with adequate seed set were propagated as Cross 30, according to the procedure outlined in (3), and originally proposed by C. H. Goulden*. In brief the procedure used was as follows: three seeds were planted from each F_2 plant, then one plant was harvested at random, thereafter, another three seeds were planted from this single plant, and this random propagation method was repeated to the F_6 generation. At this point the seeds of the three F_6 plants were propagated to produce the F_7 and subsequent generations. The cross was repeated in the field in 1950 and the F_1 was grown in the greenhouse during the winter. The F_1 (85 plants) of this cross segregated for head type similar to a backcross, and for this reason this hybrid was designated Cross 81. The seeds of single F_1 heads (about 10 seeds per head) were grown in the F_2 and subsequently each plant was propagated as described for Cross 30. Later comparisons made between lines with different segregating behaviour for head type and for rust resistance confirmed the heterozygous nature of the R.L.2265-46 parent used in Cross 81.

The first quality studies of the parents and 397 random F_6 lines were made at Ottawa in 1956. Each line was grown in a single rod row plot and the parents were grown at 20-row intervals. In 1958, a replicated test was studied using 100 F_6 random lines, each replicated four times in single rod row plots. These provided information on quality and also on yield.

The test for seedling reaction to rust was made on 200 F_6 lines at the Research Station, Canada Department of Agriculture, Winnipeg, Manitoba. A few lines were still segregating for resistance to stem rust in F_6 and in cases where less than 50 per cent of the plants of any one line were resistant these were classified as being susceptible. Similarly, lines which had less than 50 per cent compact head types were classified as fusiform.

For the quality data, 100-gram samples were conditioned to 15.5 per cent moisture and milled on a laboratory mill (2) in a uniform manner to give flour yields better than 65 per cent.

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*Personal communication.

TABLE 1.—RESULTS OF SMALL-SCALE QUALITY TESTS FOR REDMAN AND R.L.2265-46, 1958 CROP

Variety	Expansion c.c.	Sedimentation c.c.	Mixogram curve type
Redman	20.05 ± .40	53.01 ± .99	1 and 2
R.L.2265-46	13.30 ± .30	30.43 ± .97	4 only

TABLE 2.—CORRELATION COEFFICIENTS FOR EXPANSION VALUES OF 100 LINES GROWN IN 1956 AND IN 1958; AND THE MEANS FOR 200 LINES FROM THE 1956 CROP

Phenotype	Simple correlation coefficients	Mean expansion in c.c.
*Sr6*C and *Sr6*c Rust Resistant	.4008*	11.9 ± .46
*sr6*C and *sr6*c Rust Susceptible	.7038**	16.4 ± .26
*Sr6*C and *sr6*C Compact Spike	.4604**	13.5 ± .64
*Sr6*c and *sr6*c Fusiform Spike	.5876**	14.8 ± .69

*Significant at the 5% level
**Significant at the 1% level

TABLE 3.—CORRELATION COEFFICIENTS FOR EXPANSION AND SEDIMENTATION AND MEAN SEDIMENTATION VALUES OF 100 F_3 LINES, 1958 CROP

Phenotypes	Simple correlation coefficient	Mean sedimentation in c.c.
*Sr6*C and *Sr6*c Rust Resistant	.4888**	41.60 ± .67
*sr6*C and *sr6*c Rust Susceptible	.6883**	48.20 ± .80
*Sr6*C and *sr6*C Compact Spike	.5015**	40.58 ± .82
Sr6c and *sr6*c Fusiform Spike	.6190**	48.48 ± .39

RESULTS AND DISCUSSION

On the basis of the small-scale quality tests, i.e. mixograph, expansion and sedimentation, the results for twenty tests of each parental variety are given in Table 1. These data indicate that the parents differ significantly in their response to these quality tests.

To test the relationship of dough expansion values for the 1956 and 1958 crops simple correlations were calculated for 100 F_3 and F_4 lines grown in the 2 years. These correlations are recorded in Table 2 for four different phenotypes and, in addition, the mean dough expansion values are given for 200 F_3 lines from the 1956 crop. These were the only lines tested for rust reaction. The symbols *Sr6* and C refer to resistance to rust and compact spike, respectively, and *sr6* and c denote susceptibility and fusiform spike. The X^2 test of homogeneity (for the 2 years' results) is 5.235 with $P = .16$ which indicates that the different phenotypic F_3 and F_4 lines reacted in a similar manner each year. The mean expansions of the phenotypes from the 1956 crop show significant differences between the classes, with the *Sr6*C and the *Sr6*c types giving the lowest mean values and the *sr6*C and *sr6*c the highest.

The sedimentation test, which is indicative of flour strength (11), is usually significantly correlated with the expansion test*. On this premise correlations were calculated for sedimentation and expansion values of the 100 F_3 lines of the 1958 crop. These are presented in Table 3 along with the sedimentation means for each phenotype. As the X^2 value of homogeneity is 2.74 with $P = .44$, it is concluded that the different phenotypic

*Unpublished data.

TABLE 4.—CORRELATION VALUES OF EXPANSION AND SEDIMENTATION WITH MIXOGRAM CURVES FOR CROSSES 30 AND 81 AND THE MEAN VALUES FOR EACH CROSS, 1958 CROP

Cross number	Character	Simple correlation coefficients	Mean values in c.c.
30	Expansion vs. mixogram curves	.4646**	
81	Expansion vs. mixogram curves	.7429**	
30	Sedimentation vs. mixogram curves	.3216	
81	Sedimentation vs. mixogram curves	.7603**	
30	Expansion		13.8 ± .36
81	Expansion		15.6 ± .93
30	Sedimentation		40.1 ± 1.00
81	Sedimentation		49.1 ± .89

groups responded in like manner to the sedimentation test. The sedimentation mean values were lower for the groups with rust resistance and with compact spike and higher for the groups with rust susceptibility and with fusiform spike. From the mean values recorded in Tables 2 and 3 it would appear that the *Sr6* gene and the C gene, both located on chromosome XX (13), are linked with genes contributing to inferior quality as measured by these tests.

It was mentioned earlier that Cross 30 differed from Cross 81 as shown by the difference in ratio of stem rust resistant to susceptible lines and of compact to fusiform spikes. Variability in the results occurred in both crosses with respect to the expansion and sedimentation tests, but the mean values for Cross 81 were significantly higher than for Cross 30. In addition, the mixogram curves were almost entirely of a 3 or 4 type in Cross 30, whereas, in Cross 81 a distribution of all four types was observed. The correlation of the expansion and sedimentation values with the mixogram curve types, and the means of these tests for Cross 30 and Cross 81 are recorded in Table 4. These data indicate that the populations from these crosses responded differently and this was to be expected, since the rust susceptible and fusiform headed lines occurred more frequently in Cross 81 than in Cross 30.

Tables 5 and 6 show the test for independence for the frequency distribution of the mixogram curve type for rust susceptible and resistant lines and for lines with fusiform and compact spikes, respectively, for Cross 81. It is evident that both rust reaction and head type are associated with mixogram characteristics. In Table 7, six lines with a mixogram type 1 or 2 are given so as to enable a comparison with the other quality components and grain yield. The results indicate that the most promising selections are 81-263 and 81-266. These comprise 2.0 per cent of the population.

With reference to yield it may be mentioned that the average yield of 98 fusiform lines, replicated four times in single rod row plots, was 301.3 ± 4.38 grams and that of 70 compact lines was 260.8 ± 4.53 grams. The difference in mean yield is highly significant with a t value of 9.31**, indicating that the lines with compact spikes are lower in yield.

TABLE 5—TEST FOR INDEPENDENCE BETWEEN RUST RESISTANCE AND MIXOGRAM CURVE IN CROSS 81, 1956 CROP

Mixogram type	Susceptible	Resistant	Totals
1	23	2	35
2	14	6	20
3	20	11	31
4	15	15	30
Totals	82	34	116

X^2 = 16.213 and P = less than .01

TABLE 6.—TEST FOR INDEPENDENCE BETWEEN COMPACT SPIKE AND MIXOGRAM CURVE IN CROSS 81, 1956 CROP

Mixogram type	Fusiform	Compact	Totals
1	35	0	35
2	17	3	20
3	21	10	31
4	12	18	30
Totals	85	31	116

X^2 = 31.593 and P = less than .01

TABLE 7.—EXPANSION, SEDIMENTATION AND YIELD OF PARENTS OF CROSS 81 AND SIX F_9 LINES WITH STRONG MIXOGRAM CURVE TYPES, 1958 CROP

Parent or selection	Phenotype	Mixogram type	Expansion	Sedimentation	Grain yield
			c.c.	c.c.	g.
Redman	*sr6*c	1 or 2	20.0	53.0	316.0
R.L.2265-46	*Sr6*C	4	13.3	30.4	252.0
81-263	*Sr6*c	1	20.5	58.7	372.0
81-266	*Sr6*c	1	19.9	55.7	364.0
81-132	*Sr6*c	2	16.2	54.5	372.0
81-46	*sr6*C	2	20.7	62.1	282.0
81-99	*Sr6*C	2	20.8	45.9	198.0
81-202	*Sr6*C	2	17.9	54.4	194.0

Inheritance of Stem Rust Resistance and Compact Spike in Cross 30 and Cross 81

The data given in Table 8 show that stem rust resistance and head density are controlled by single genes. They reveal that the genotype of the R.L.2265.46 plant used in Cross 30 was *Sr6Sr6*CC. Since ratios of 1:3 were observed in advanced F_9 lines of Cross 81 for rust reaction and head density it appears that the parent used in this cross was of the genotype *Sr6sr6*Cc. The X^2 for the goodness of fit tests, recorded in Tables 9 and 10, respectively, for the *Sr6* and C genes of F_9 lines gave P values of between .02 to .05 for Cross 30 and less than .01 for Cross 81. The reason for these

TABLE 8.—GOODNESS OF FIT TO SINGLE GENE RATIOS FOR STEM RUST RESISTANCE AND SPIKE TYPE IN 84 F_3 LINES OF CROSS 30 AND 116 LINES OF CROSS 81, 1958 CROP

Cross	Phenotypes	Ratio	X^2	P—values
30	*Sr6*, 37 : *sr6*, 47	1 : 1	1.190	.20 to .50
30	C, 44 : c, 40	1 : 1	.762	.20 to .50
81	*Sr6*, 34 : *sr6*, 82	1 : 3	1.149	.20 to .50
81	C, 31 : c, 85	1 : 3	.184	.50 to .95

TABLE 9.—GOODNESS OF FIT TO A 1:1:1:1 RATIO OF STEM RUST RESISTANCE AND HEAD TYPE IN 84 F_3 LINES OF CROSS 30, 1958 CROP

Phenotypes	Observed	Calculated	O-C	$\frac{(O-C)^2}{C}$	
*Sr6*C	24	21	3	.428	
*Sr6*c	13	21	−8	3.047	
*sr6*C	16	21	−5	1.190	
*sr6*c	31	21	10	4.762	
Total	84	84	0.0	8.427	P = .02 to .05

TABLE 10.—GOODNESS OF FIT TO A 1:3:3:9 RATIO OF STEM RUST RESISTANCE AND HEAD TYPE IN 116 F_3 LINES OF CROSS 81, 1958 CROP

Phenotypes	Observed	Calculated	O-C	$\frac{(O-C)^2}{C}$
*Sr6*C	22	7.25	−14.25	2.801
*Sr6*c	12	21.75	9.75	4.371
*sr6*C	9	21.75	12.25	6.899
sr6c	73	65.25	−7.75	.921
Total	116	116.00	0.0	14.992

P = less than .01

low P values is probably due to linkage. Peterson and Campbell (12) suggested that the genes for rust resistance and head density in McMurachy are linked with a crossover value of "about 30 per cent". Knott (7) reported that the genes *Sr6* and C are linked and that the recombination value is 18.1 per cent. A linkage value was calculated for both crosses in this study. For Cross 30 this value is 33.1 ± .7 per cent and in Cross 81 the recombination is 18.9 ± 0.3 per cent.

SUMMARY AND CONCLUSIONS

Flour quality, stem rust resistance to race 15B-4 (Can.) controlled by the gene *Sr6*, and compact spike were studied in unselected, random F_3 and F_3 lines of two crosses involving two phenotypically similar derivatives of a hybrid strain, R.L.2265-46 (McMurachy X Exchange) with Redman.

Components of quality were evaluated by the expansion, sedimentation and the mixograph tests. In 1956, 397 F_3 lines, grown in single plots, were used for the mixograph and expansion quality tests. The 1958 studies in-

cluded, in addition, sedimentation quality data and data on grain yields. However, only 100 F_6 random lines grown in single rod row plots with four replications were used in 1958 for quality tests.

The interannual simple correlation coefficients between the expansion tests in four different phenotypic groups of F_5 and F_6 lines were significant and homogenous. Similarly, significant correlations were obtained between the expansion and sedimentation values of the above groups of F_6 lines. The means for the quality tests show that the rust-resistant group of lines and the group of lines with compact spikes are significantly lower than those having the contrasting characters. The X^2 for Independence between stem rust reaction, head density and mixogram evaluation indicates a close association between these characteristics and curve type. Of the 100 F_6 lines evaluated in 1958, approximately 2.0 per cent were equal in quality and yield to Redman and in addition were resistant to race 15B-4 (Can.). The reason for the inferior quality lines obtained in Cross 30 as compared to Cross 81 cannot be explained by the *Sr6*C combination of genes alone. It is possible that the plants of the R.L.2265.46 parent used in these crosses differed also in quality.

The mean yield of the fusiform lines was significantly higher than that of lines having compact spikes. Rust resistance and compact spike were found to be conditioned by single genes which failed to segregate independently. Crossover values of 33.1 ± 0.7 per cent for Cross 30 and 18.9 ± 0.3 per cent for Cross 81 were obtained.

One may conclude from this study that where dominant major genes are linked and at least one of these is associated with poor quality it may be more efficient to use the corresponding recessive more than once in the crossing program in order to obtain sufficient desirable combinations among the lines of a population. This, of course, may be most readily achieved by the backcross method.

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COMPARISON OF LIGHTLY GRAZED AND UNGRAZED RANGE IN THE FESCUE GRASSLAND OF SOUTHWESTERN ALBERTA[1]

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ABSTRACT

The influence of grazing on the vegetative cover of fescue grassland in southwestern Alberta was assessed by studying two adjoining sites, one lightly grazed, the other ungrazed. Percentage basal area, yield, water-intake rate, soil temperature, soil moisture, and amount of root material were compared on a paired plot basis.

The data showed that light grazing resulted in the development of a richer flora dominated by *Danthonia parryi*. Protection from grazing appeared to simplify the flora with a trend toward a cover consisting largely of *Festuca scabrella*. There was little evidence of difference in productivity between the two sites. Cooler and moister conditions prevailed in the upper 12 inches of the soil profile of the ungrazed site as a result of heavy accumulation of mulch. Considerably more root material to a depth of 54 inches was present on the lightly grazed site. The harmful effects of herbage removal, shown by clipping studies, were not apparent in the field study under a light rate of grazing.

INTRODUCTION

Range management literature stresses the harmful effects of grazing, as the tendency is to describe regressive trends from protected to overgrazed sites. Consequently, little is known about grazing pressure as an ecological influence (3).

Light grazing is frequently suggested as a means of range improvement and studies have shown the practice to be feasible (4, 5, 10). This is at variance with much of the experimental evidence which suggests that herbage removal is damaging. Too little is known of the effects of light grazing to permit any very meaningful conclusions (3).

The objectives of the study reported herein were to obtain quantitative data on the differences between lightly grazed and ungrazed range sites and to assess the effects of grazing *per se* on the vegetative cover.

DESCRIPTION OF THE AREA

At the Range Experiment Substation, Stavely, Alberta, an area that had been lightly grazed (average utilization 15 to 25 per cent) for a period of 12 years was available for study. A 2-acre exclosure within this field and an adjoining 2-acre plot from the field itself were selected for comparison (Figure 1).

From 1884 to 1908 the site was grazed by cattle on a moderate use basis (1). From 1908 to 1920, the area carried horses again on a moderate summer use basis. It was used as summer range for cattle from 1920 to 1943 and was heavily grazed during the drought years of the thirties. In 1944 the area became part of a large ranch and was lightly used as winter pasture until 1949. In 1949 the area was lightly stocked with cattle on a summer use basis; the exclosure was fenced and protected from all grazing by livestock. This type of use was maintained until 1960.

[1]Contribution from the Forage Crops Section, Canada Agriculture Research Station, Lethbridge, Alta.

The vegetation of the area has been described by Moss (7) and Moss and Campbell (9). It is considered part of the *Festuca scabrella* association, although it has a richer flora than much of the remainder of the association. *Danthonia parryi* and *Festuca idahoensis* are the chief associated grasses. *D. parryi* is of particular interest since it behaves as an edaphic subclimax species on exposed sites and as a disclimax under heavy utilization (8).

The study site was situated on orthic shallow black soil developed on glacial till. The texture of the soil ranged from loam to clay loam. The climate has been classified as "moist subhumid" with little or no water deficiency at any season (12). Average annual precipitation near the study sites for the period 1950-1959 was 23.91 inches of which 9.21 inches, or approximately 40 per cent, was received as snow.

METHODS

The study was divided into three parts as follows:

1. *Vegetation*

A vegetation analysis was conducted by the vertical point method (6). Three thousand points per study site were examined and the percentage basal area was determined in August, 1960. Basal area data obtained by the same method in August, 1949, were available for comparison.

Yields of the various components of the vegetative cover were determined in late July, 1960, by clipping 30 four-square-foot plots on each study area. The clipped material was divided into green grass, green forbs and shrubs, fresh mulch, and humic mulch according to the method of Dyksterhuis and Schmutz (2). Green grass consisted of all live portions and the dead tips of the growing grasses; *Carex* spp. were included in this group. Green forbs and shrubs consisted of the living, above-ground portion of these plants and included partially dead stems. Fresh mulch consisted of the fresh residuum of herbage and was made up of the upper layer of bulky, coarse, leafy, and largely undecayed natural mulch. Humic mulch consisted of largely decayed, disintegrated, and fragmented organic residuum of fresh mulch. The dry matter contents of these materials were determined.

Plant vigour was rated by measuring the maximum leaf height and average basal diameter of 40 paired plants of *F. scabrella*.

2. *Soil*

Soil moisture samples were taken at 12-inch increments to a depth of 48 inches with a King tube. Thirty cores were obtained in each study area. Soil moisture was determined by oven drying the samples for 24 hours at 220°F. Soil temperatures at an 8-inch depth were recorded daily at ten paired locations from August 1 to October 31, 1960.

A measure of water-intake was obtained with a mobile infiltrometer according to the procedure outlined by Rauzi (11). A simulated rainfall intensity of 3.60 inches per hour was applied to three 4-square-foot plots in each study area. Yields of herbage and mulch and soil loss were determined on the same plots.

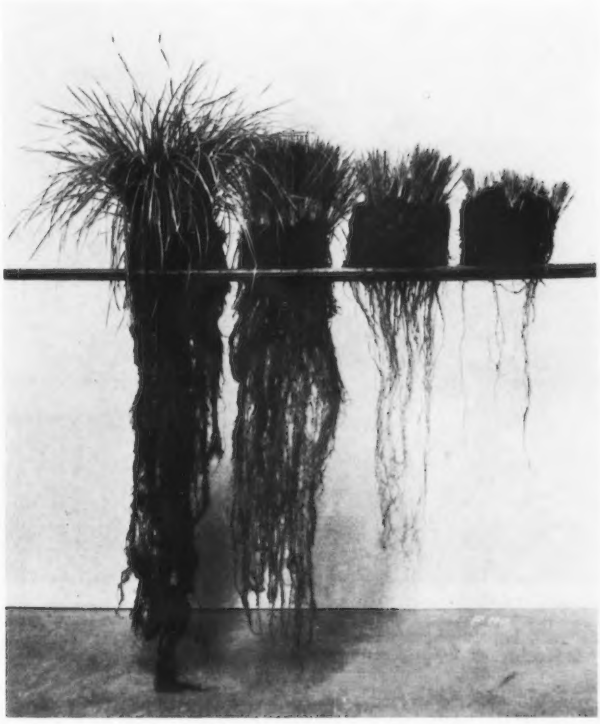

FIGURE 2. Plants of *Festuca scabrella* unclipped and clipped to stubble heights of 5 inches, 3 inches, and 1½ inches at 4-week intervals.

FIGURE 1. General view of the study sites showing the ungrazed exclosure (*broken line*) and lightly grazed area (*solid line*). The study sites were located on a moderate west-facing slope.

Thirty 2½-inch cores were obtained by 6-inch increments to a depth of 54 inches within each study area. Since the samples were found to be too small for effective root washing, the separate increments for each treatment were bulked, washed, and a single weight of roots recorded.

Student's Pairing Method was used in the analysis of part of the data obtained in (*1*) and (*2*) above.

3. Greenhouse Study

Sods containing single plants of *F. scabrella* were removed from an ungrazed stand, trimmed to 6 x 6 x 4 inches, and transplanted into metal containers filled with a 60:40 mixture of loam and sand. The containers measured 10 x 10 x 28 inches deep and had one removable side. Top-growth was removed when the sods were placed in the greenhouse. Tiller numbers of all plants were made approximately equal at the beginning of the clipping experiment and were determined at the end of the 16-week growing period. Four clipping intensities were used; clipped at the end of 16 weeks and at 4-week intervals to stubble heights of 1½, 3, and 5 inches. Yields of top-growth and root-growth were determined at the end of the growing period.

RESULTS

The percentage basal area of vegetation on the lightly grazed and ungrazed sites in 1960 and the percentage basal area in 1949 are presented in Table 1. *D. parryi* decreased under protection while *F. scabrella* increased. This would seem to support Moss (8), who stated that the former grass behaves as a disclimax under grazing pressure. The basal area of *D. parryi* was maintained under light grazing even with the increase in basal area of *F. scabrella*. *F. idahoensis*, which has reacted as an increaser under heavy grazing, showed a slight reduction in percentage basal area under light grazing. Percentage basal area of *Bromus pumpellianus* increased considerably from 1949 to 1960 on the ungrazed site and to a lesser extent under light grazing. The increased growth occurred between the tussocks of *F. scabrella* and in the presence of natural mulch. The increased percentage basal area of "Forbs and shrubs" shown for both the ungrazed and lightly grazed sites was due to an increase in size of plants of *Potentilla fruticosa*.

TABLE 1.—AVERAGE PERCENTAGE BASAL AREA OF VEGETATION OF TWO STUDY SITES AT STAVELY IN 1949 AND 1960

Species[1]	Study sites in 1949	Ungrazed 1960	Lightly grazed 1960
Danthonia parryi	8.25	5.57	8.80
Festuca scabrella	3.75	6.03	4.97
Festuca idahoensis	1.35	1.53	1.10
Agropyron subsecundum	0.35	0.13	0.40
Bromus pumpellianus	0.01	1.40	0.60
Other grasses	0.69	0.07	0.40
Carex spp.	1.80	1.50	2.77
Forbs and shrubs	3.30	4.29	4.74
Total	19.50	20.52	23.78

[1]Nomenclature follows that of Moss, E. H. Flora of Alberta. Univ. Toronto Press. 1959.

TABLE 2.—HEIGHT AND DIAMETER OF FESTUCA SCABRELLA PLANTS AND YIELDS OF SOME COMPONENTS OF THE VEGETATIVE COVER, STAVELY, 1960

	Ungrazed	Lightly grazed	D.F.	"t"
Festuca scabrella:				
Average plant height (inches)	19.8	19.0	39	1.11
Average plant diameter (inches)	4.39	3.38	39	2.46*
Yield (pounds dry matter per acre):				
Green grass	1,464	1,252	29	2.09*
Green forbs and shrubs	762	936	29	1.24
Total green herbage	2,226	2,188	29	0.28
Fresh mulch	3,889	2,881	29	4.00**
Humic mulch	7.154	4,850	29	4.94**
Total mulch	11,043	7,731	29	5.70**
Total organic matter	13,299	9,919		

*Significant at P 0.05
**Significant at P 0.01

Total number of species detected by the point method, including *Carex* spp., was considerably greater under light grazing (31 species) than on the protected area (19 species) or on the study sites in 1949 (23 species). Nine grass species were detected in 1949, while in 1960 eight species were noted on the lightly grazed area and six species on the ungrazed site. Thirteen species of forbs and shrubs were detected in 1949, while twenty-two were noted under light grazing and twelve under protection in 1960. The additional forbs detected under light grazing included *Thalictrum venulosum*, *Heuchera richardsonii*, *Zizia aptera*, *Anemone patens* var. *wolfgangiania*, *Taraxacum officinale*, *Agoseris glauca*, *Lupinus argenteus*, *Geum triflorum*, *Fragaria virginiana*, and *Thermopsis rhombifolia*. *Stipa spartea* var. *curtiseta* and *Agropyron dasystachyum* were detected in 1949 but were not recorded on the protected site in 1960.

Comparisons of several constituents of the vegetative cover are shown in Table 2. Plant vigour, as reflected by the height of *F. scabrella*, did not differ between treatments. Diameter of plants was significantly different (P 0.05), smaller plants being characteristic of the lightly grazed area. This is in agreement with Moss and Campbell (9), who pointed out that a decrease in diameter of tussocks occurs when fescue prairie is utilized.

The amount of surficial organic matter found on the study sites (Table 2) was comparable to that reported for tall-grass prairie by Dyksterhuis and Schmutz (2). The differences between the two sites were principally in the amounts of fresh and humic mulch which were higher (P 0.01) on the ungrazed site. Differences in yield of green herbage on the two sites were not significant and the apparent difference in grass yields was probably caused by grazing. The lightly grazed study site was accessible to cattle from May 15 to July 20, 1960.

Cooler and moister conditions prevailed in the upper 12 inches of the soil profile under protection than on the lightly grazed site (Table 3). Soil temperature differences were small and tended to become equal as the season advanced. Percentage soil moisture was higher (P 0.05) in the

TABLE 3.—MEAN MONTHLY SOIL TEMPERATURES AT AN 8-INCH DEPTH ON TWO STUDY SITES

Month	Ungrazed	Lightly grazed
August, 1960	52°F.	54°F.
September, 1960	48°F.	49°F.
October, 1960	43°F.	43°F.

TABLE 4.—PERCENTAGE SOIL MOISTURE AT VARIOUS DEPTHS ON TWO STUDY SITES

Soil depth	Ungrazed	Lightly grazed
0-12 inches	22.19	19.38*
12-24 inches	14.49	14.35
24-36 inches	13.34	13.52
36-48 inches	12.75	14.00

*Significant at P 0.05

TABLE 5.—WEIGHT OF UNDERGROUND PLANT MATERIAL IN POUNDS PER ACRE AT VARIOUS DEPTHS ON TWO STUDY SITES

Soil depth	Ungrazed	Lightly grazed
0- 6 inches	11,193	20,067
6-12 inches	1,109	3,297
12-18 inches	1,797	2,293
18-24 inches	884	1,520
24-30 inches	333	688
30-36 inches	370	470
36-42 inches	351	407
42-48 inches	170	281
48-54 inches	218	242
Total	16,425	29,265

upper 12 inches on the ungrazed site but did not differ between sites at depths of from 12 to 48 inches (Table 4).

Dry weights of root material at various depths from the two sites are shown in Table 5. It is apparent that more root growth occurred on the lightly grazed area than on the protected site at all depths.

Water-intake rate and soil loss are shown in Table 6. Average values for the second 30-minute period and the fourth 15-minute period of a 1-hour run indicate that increasing amounts of natural mulch on the protected site (Table 2) resulted in greater water-intake rates and in lower soil loss per acre. The water application intensity of 3.60 inches per hour used in this test was probably well in excess of natural rainfall intensity for the area.

Results of the greenhouse clipping experiment indicated that herbage removal was damaging to the individual range plant. Four plants of *F. scabrella*, one unclipped, the others clipped to stubble heights of 5, 3, and

TABLE 6.—WATER-INTAKE RATE AND SOIL LOSS FROM TWO STUDY SITES

	Ungrazed	Lightly grazed
Water-intake rate during 1-hour run of:		
Second 30-minutes (inches/hour)	2.24	1.65
Fourth 15 minutes (inches/hour)	2.20	1.59
Soil loss (pounds per acre)	65	133

1½ inches at 4-week intervals, are shown in Figure 2. The 5-inch clipping treatment, comparable to 20 per cent utilization, resulted in a considerable reduction in weight of roots as compared to the unclipped control (Table 7). Tiller production was vigorous when plants were clipped to a 5-inch stubble height but was reduced when plants were clipped to 3 inches. Comparable results have been obtained in greenhouse clipping studies with *F. idahoensis* and *D. parryi**.

DISCUSSION

The character of the vegetation on the study sites was changed as a result of the treatments imposed during the 12-year period. Light grazing tended to favour dominance by *D. parryi* while protection from all grazing favoured dominance by *F. scabrella*. Light grazing resulted in the development of a more varied flora with a greater total percentage basal area while protection from grazing appeared to simplify the flora with a trend toward a cover consisting largely of *F. scabrella*. A varied cover could conceivably be of value to the ecosystem especially during times of climatic stress. The change in character of the vegetation was not accompanied by a change in production. In terms of percentage basal area, there was little doubt that an improvement in range condition had occurred during the 12 years of light grazing.

Large amounts of natural mulch had accumulated on the ungrazed site and seemed to be partially responsible for the simplification in plant composition that had taken place under this treatment. The importance of

*Unpublished data

TABLE 7.—EFFECT OF VARIOUS CLIPPING TREATMENTS ON FESTUCA SCABRELLA PLANTS IN THE GREENHOUSE

Treatment	Yield of dry matters in grams		Number of tillers		Estimated percentage utilization
	Top-growth	Root-growth	Initial	Final	
Unclipped	20.16	15.03	87	431	0
Clipped every 4 weeks to 1½ inches	1.84	0.68	73	53	90
Clipped every 4 weeks to 3 inches	5.91	2.98	81	192	70
Clipped every 4 weeks to 5 inches	15.96	7.83	77	427	20

natural mulch in conditioning the soil surface for infiltration and resistance to erosion was shown.

The weight of the root mass was greater under light grazing than under protection. Similar results have been noted elsewhere (14). Troughton (14) states that results from a clipping experiment with *Lolium perenne* showed that the weight of roots per plant decreased with increasing intensity of defoliation but that the root weight per unit area of sward increased. This was attributed to an increase in plant density as a result of defoliation. Stevenson and White (13) have also reported a greater weight of roots under grazed than under ungrazed prairie. Thus the increased percentage basal area found under light grazing accounts in part for the difference. Another reason for this difference may be related to a factor that was not assessed in this study. Troughton (14) reports that a number of investigators have noted reduced root growth as a result of partial shading. Shading was a greater factor on the ungrazed site as a result of dominance by a tall-growing species and large amounts of mulch than on the lightly grazed area.

Results obtained from clipping studies in the greenhouse appeared to contradict those obtained in the field. In the greenhouse, a considerable reduction was noted in amount of top- and root-growth when 20 per cent of the herbage was removed. The field study, after 12 years of comparable utilization, showed an increase in amount of root-growth and a slight reduction in amount of top-growth, the latter being attributed to prior utilization by cattle. Wilson (15) reported similar contradictory results between greenhouse and field clipping tests with *F. rubra* and *Dactylis glomerata*. In the greenhouse, yields of both top- and root-growth declined as the severity of clipping increased while, in the field, higher yields were obtained by clipping to a lower stubble height. In a subsequent greenhouse experiment, Wilson (15) used larger sods, harvested the centre portion only, and duplicated field results. He considered that a high light intensity near the base of the plant was important to regrowth and concluded that this was available to potted plants in the greenhouse under all stubble heights but only to closely clipped plants in the field because of self-shading. A similar explanation may apply to the study reported herein. The data suggest that greenhouse clipping tests should be used with caution when applied to actual grazing studies.

CONCLUSIONS

A light rate of grazing on the study site resulted in some diversification in plant composition, a greater total percentage basal area, a lesser accumulation of natural mulch, and in a greater amount of underground plant material per unit area. The harmful effects of herbage removal shown by a clipping experiment were not apparent under a light rate of grazing. This suggests that any beneficial effects of grazing accrue to the ecosystem.

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INDUCTION OF SEED SET IN DWARF-SPRING AND WINTER WHEAT

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ABSTRACT

Studies were made of the effect of gibberellin (GA) application to dwarf plants of spring wheat (*Triticum aestivum* L.) which resulted from crossing monosomic and disomic Redman with Kenya Farmer. GA at 10 and 100 p.p.m. induced the formation of 53 seeds on 12 plants sprayed 46 days after planting. Fewer seeds were produced on a larger number of plants sprayed at a later stage. The application of IAA, NAA and TIBA to dwarfs at 5 p.p.m. was ineffective in increasing stem length or inducing heading. Cold-shock treatment of seeds appeared to induce heading in one experiment and not in another. GA did not affect the course of meiosis but it did cause deformities in plants grown from treated seed. Deformities included branching of the main stem, development of crown roots from two or three nodes per stem, and rapid growth of a "needle-like" internode. Application of fertilizer overcame yellowing but not the spindly growth which occurred in GA-treated plants. Two winter wheat varieties were given GA and cold treatment both of which induced heading in one variety and not in the other. GA induced extensive growth of leaves and stems in both winter wheats under low intensity light and high temperature whereas under opposite conditions it caused retarded growth and delayed maturity.

INTRODUCTION

The work of plant breeders, attempting to transfer rust resistance in wheat, has been hampered by dwarfing. Some success has been reported in overcoming the dwarf habit in certain crops by applying chemical growth substances. This paper reports on the use of gibberellin (hereinafter referred to as GA) and other chemicals as well as cold shock treatment in an attempt to overcome dwarfing or induce heading and seed set in dwarf plants.

Several reviews of the GA work are available (2, 3, 8, 9). Practically every application of GA, where penetration to the growing parts has been accomplished, resulted in rapid growth and elongation. The chemical is systemic and highly mobile, but its effect is only temporary, making repeated applications necessary. Both cell elongation and cell division is promoted in above-ground parts, but in some reports root growth has been shown to be unaffected or retarded. New growth is usually a pale yellow-green colour, a condition which can be corrected by maintaining a high level of fertility and optimum growing conditions or by applying sucrose solution as a foliar spray.

Considerable attention has been given to the problem of overcoming dwarfing caused by genetic, physiological or disease factors in various plant groups (1, 2, 3, 6). Some treated dwarfs in corn and apple have attained normal height when sprayed with GA. This suggests a biochemical basis for heritable characteristics (9). Allan *et al.* (1), in studying the effect of GA on dwarfing in winter wheat, reported that 100 p.p.m. of GA produced a greater stimulation than rates of 1, 10 or 1000 p.p.m. Growth regulators other than GA have also been used in attempts to induce flowering.

Contrary to the results of some cereal crop investigators, Caso *et al.* (4) found that, under certain circumstances, GA did induce earlier flowering in fall rye. While early applications inhibited flowering, GA treatment of plants having 10 exposed leaves caused the 20th to the 25th primordia, which are labile, to form flowers rather than leaves. Since GA occurs naturally in many plants, including wheat (7), and since it induces extension of growth, it might be expected that a dwarf plant is lacking in either GA or a precursor to it.

MATERIAL AND METHODS

In a genetic study the 21 Redman monosomics and diploid Redman were crossed to Kenya Farmer. The F_1s from the two crosses, hereinafter referred to as RM x KF and R x KF, were planted. Line VIII and line XIII from RM x KF segregated for tall and dwarf habit (Figure 2) as chromosomes VIII and XIII of Redman carry complementary dominant genes for dwarf habit (5). A few F_1s from R x KF were also normal, probably because of heterozygosity for dwarfing genes in the parents. All other F_1 plants from both crosses were dwarf. Applications of 16-20-0 fertilizer was made monthly in all experiments. Fifteen pots containing dormant seed were placed outside for 48 hours during which time the temperature went down to –4°C. Ten of the fifteen seeds grew and six of them produced plants of normal growth habit.

Experiment I

In a preliminary attempt to induce dwarf plants to set seed, the following chemical treatments were applied to four single-plant replicates of the F_1s of R x KF 46 days after planting:

1.	Indoleacetic acid (IAA)	5 p.p.m.
2.	2,3,5,-triiodobenzoic acid (TIBA)	5 p.p.m.
3.	Naphthalene acetic acid (NAA)	5 p.p.m.
4.	Gibberellic acid (GA)	10 p.p.m.
5, 6.	Gibberellic acid	100 p.p.m.

Treatments were supplied in six foliar applications at 3-day intervals, except Treatment 6, which was applied by placing drops in the axils of leaves nine times at 2-day intervals. Plants receiving drops remained on the greenhouse bench, but plants receiving spray treatments were placed in a plastic-walled incubation cabinet over night. The humidity was kept high enough so that the droplets of the spray remained on the leaves for 8 to 10 hours. Two height measurements, i.e. average height of leaf auricle and average length of leaf, were taken on each plant at the time of application and at three later stages.

Experiment II

Because GA gave the most promising results in Experiment I, it was used in succeeding tests. All dwarf F_1s from RM x KF, which were still growing in the greenhouse, were reduced in number of tillers and sprayed twice with 50 p.p.m. of GA, the interval between spraying being 1 week. These plants had been sown 75 days previously and had produced from 25 to 50 tillers each. The number of tillers was reduced to 4, 5 or 6 in the

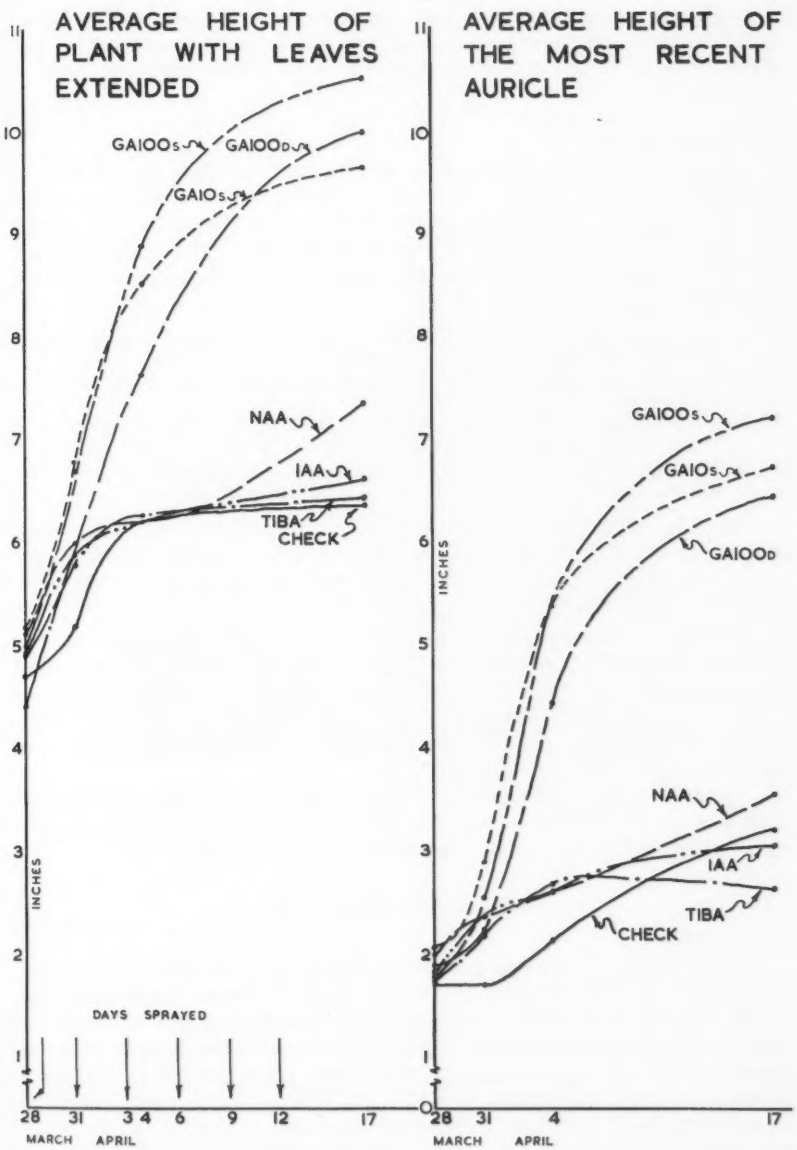

FIGURE 1. Plant heights in inches at various stages following treatment with GA at 10 and 100 p.p.m. applied by spraying plants (GA10S and GA100S), at 100 p.p.m. applied by placing drops in leaf axils (GA100D), and NAA, IAA and TIBA sprayed on plants at 5 p.p.m. Arrows indicate dates for spray application. Measurements were made in inches from soil surface.

hope that stronger culms and more vigorous heads would produce more seed. In this experiment soil fertility was maintained by substituting two applications of nutrient solution for one 16-20-0 treatment just prior to heading.

Experiment III

Since these plants did set some seed in spite of their age at time of treatment, a new set of RM x KF and R x KF F_1s were subjected to a number of treatments. In addition to the Redman and Kenya Farmer checks, one easy-(IBO 1535) and one difficult-to-vernalize (Zanda) winter wheat was added to check the effect of the various treatments on winter wheat. Treatments were as follows:

1. Seeds germinated in winter, planted and the plants sprayed with 100 p.p.m. of GA weekly from emergence to heading.
2. Seeds germinated for 24 hours in GA, transferred to water for completion of germination, planted and sprayed as in 1.
3. Seeds germinated as in 2, subjected to temperatures of −2°C. to −6°C. for 10 hours, planted and sprayed as in 1.
4. Seeds germinated in water and cold treated as in 3 (no GA).
5. No treatment (R x KF and Redman and Kenya Farmer only).

No direct check for the RM x KF material was used because of shortage of seed but R x KF was used to check both crosses. Checks for the winter wheats were unnecessary since no heads could be produced in the length of time of these experiments.

Because of the large number of plants in this experiment, spray was applied to plants on the greenhouse bench. High humidity was maintained by spraying the floor and walls with water at the time of application and again two hours later. Plants in Experiment III were grown under conditions of an 18-hour day, low light intensity and moderately high temperatures (day 70-90°F., night 65-70°F.).

In addition IBO 1535 and Zanda were germinated, planted and sprayed with GA as in Treatment 1 above but grown under conditions of low temperature (62 ± 3°F.) and continuous light of high intensity.

RESULTS AND DISCUSSION

Experiment I

In Experiment I, GA had a much greater influence on dwarf wheat than IAA, TIBA or NAA (Figure 1). Leaves on GA treated plants continued to grow much more rapidly than those of the check plants while plants treated with the other chemicals appeared to be stimulated slightly with one application but not with subsequent treatments. The growth of the check plants soon caught up to that of IAA- and TIBA-treated plants but those treated with NAA maintained their lead in growth. The 100 p.p.m. spray application of GA gave only a very slight increase over GA applied at 10 p.p.m. The application of drops in the axils of leaves was slightly less effective than the foliage spray. This was probably due to less leaf area for absorption or because the drops soon evaporated. In spite of the more frequent treatment less GA was actually taken into the plant.

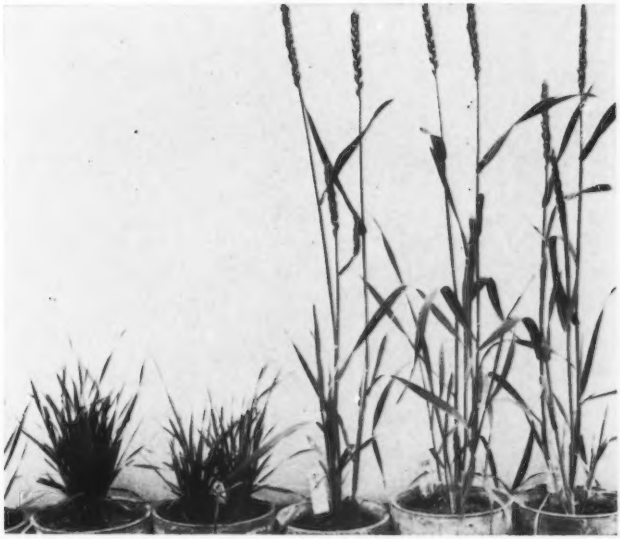

FIGURE 2. Dwarf and tall F_1 plants from Redman x Kenya Farmer showing stages when sprayed with gibberellic acid.

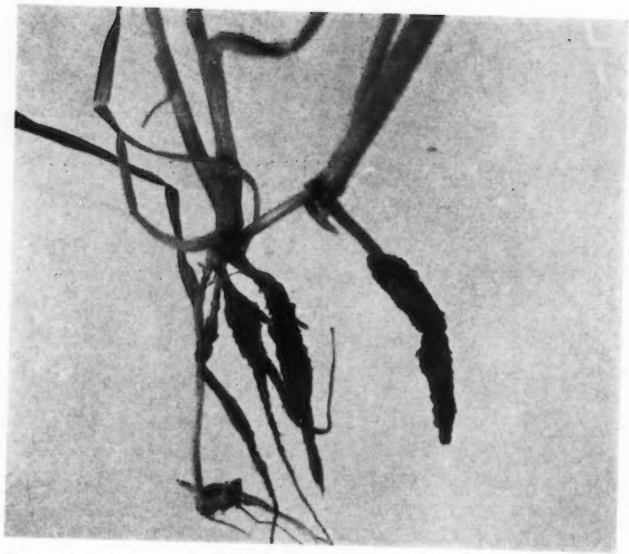

FIGURE 3. A GA-sprayed winter-wheat plant which has established a second crown.

TABLE 1.—NUMBER OF HEADS AND SEEDS PRODUCED BY F_1 DWARFS WHEN TREATED WITH CHEMICALS (experiments I and II)

Material treated	Treatment	No. of plants treated	No. of heads	No. of seeds
R x KF 46-day-old plants	GA 10 p.p.m. sprayed	4	2	8
	GA 100 p.p.m. sprayed	4	4	19
	GA 100 p.p.m. drops	4	2	26
	IAA 5 p.p.m. sprayed	4	1	19
	NAA 5 p.p.m. sprayed	4	0	0
	TIBA 5 p.p.m. sprayed	4	0	0
	Check	4	0	0
RM x KF 75-day-old plants	GA 50 p.p.m. sprayed	81	9	34
	Check	100	0	0

Analysis of variance of the data on leaf and auricle height indicated that the differences were nearly significant at the 5 per cent probability level 3 days after the first treatment. At 7 and 20 days following application GA-treated plants showed significant differences at the 1 per cent level.

As shown in Table 1, 12 GA-treated plants produced 53 seeds. The 19 seeds produced on one head of one of the IAA-treated plants were unexplainable since no sign of heading occurred on any other tiller of the four plants.

Experiment II

In Experiment II the 81 dwarf F_1 plants from RM x KF sprayed with GA when 75 days old produced 34 seeds (Table 1). When these results are compared with Experiment I it appears that: (a) application to vigorously growing plants was more effective than to older plants, and (b) GA could be used with some success to induce heading and seed set in dwarf wheat.

Many spikes on the GA-treated plants had a high percentage of sterile florets but pollen-mother-cell analyses revealed no abnormalities. Sterility appeared to be due to the spindly condition of the stems and spikes which developed in spite of adequate soil fertility.

Experiment III

Because of the findings of these two experiments and the apparent induction of normal growth habit by cold-shock treatment the third experiment summarized in Table 2 was conducted. Mortality was high when cold-shock treatment was applied to germinating seed but this treatment induced heading in 6 of the 25 F_1 plants which survived. More of the seeds of Kenya Farmer and IBO 1535 survived the cold treatment than of Redman

TABLE 2.—HEADING INDUCTION EFFECTS OF GA AS A SEED TREATMENT AND AS A FOLIAGE SPRAY WITH AND WITHOUT COLD-SHOCK TREATMENT OF GERMINATING SEED IN THE DWARF F_1 OF RM X KF AND R X KF AND IN REDMAN (R), KENYA FARMER (KF), ZANDA (Z) AND IBO 1535 (IBO)

Treatment	Materials treated	Dwarf F_1		Parents Winter wheat			
		RM x KF	R x KF	R	KF	Z	IBO
GA 100 p.p.m. spray	No. of seeds treated	57	10	4	4	12	12
	No. of plants grown	49	10	4	0	7	9
	No. of plants headed	44	10	4	—	—	8
	Days to head	75	74	50	—	—	51
GA seed treatment GA spray	No. of seeds treated	54	10	0	0	0	0
	No. of plants grown	43	10	—	—	—	—
	No. of plants headed	21	9	—	—	—	—
	Days to head	76	76	—	—	—	—
GA seed treatment GA spray cold-shock treatment	No. of seeds treated	57	10	4	4	12	12
	No. of plants grown	29	8	4	2	6	6
	No. of plants headed	11	5	4	2	—	6
	Days to head	71	69	40	44	—	51
Cold-shock treatment	No. of seeds treated	38	10	4	4	12	12
	No. of plants grown	25	2	0	4	2	10
	No. of plants headed	6	0	—	4	—	10
	Days to head	85	—	—	61	—	56
Check (no treatment)	No. of seeds treated	0	19	4	4	0	0
	No. of plants grown	—	19	4	2	—	—
	No. of plants headed	—	0	4	2	—	—
	Days to head	—	—	47	53	—	—

and Zanda. Cold-treated Kenya Farmer was later heading than the Kenya Farmer check. The cold-shock treatment vernalized IBO 1535 as all plants headed in an average of 56 days. In this variety cold treatment in combination with GA seed treatments and foliage applications appeared to speed up head development by 4 or 5 days compared to GA applications with no cold treatment.

GA applied weekly induced almost all dwarf plants to head but these plants did not exceed 14 inches in height. This treatment did not shorten the time to head for Redman but did decrease by 5 days the time required for IBO 1535 to head when compared with cold-treated plants. By comparison germination of seeds in GA solution resulted in many subsequent plant deformities and thus a decrease in the degree of heading in dwarfs from the RM x KF cross (Table 2).

Deformities included branching and the formation of crown roots at nodes above the ground (Figure 3). In several plants the stems grew very rapidly after GA application and had spindly, "needle-like" internodes ranging from one to four inches long. These needle-like sections of the stems supported considerable growth and branching in some plants. Sometimes this section buckled as it grew and forced its way through the leaf sheath to form a semi-circular bow in the stem. When this happened the

leaf sheath helped support the plant. Fewer deformities occurred in the R x KF cross which suggests that the RM x KF dwarfs were less resistant to abnormalities caused by GA seed treatment. GA replaced the cold requirement of IBO 1535, the easy-to-vernalize winter wheat, but 86 days after planting Zanda showed no signs of heading.

In spite of the many heads which were produced on the dwarf plants very little seed development occurred in this test. This was not surprising because the tall plants from line VIII and the parents produced only five or six seeds on plants with two or three heads. The reason for poor seed set was attributed to excessive day temperatures during seed setting time. Dwarf plants which produce weak, spindly heads when grown under good growing conditions would not be expected to set seed under more adverse conditions.

Light and temperature appeared to influence the effect of GA treatment on the winter wheat varieties, IBO 1535 and Zanda, but not the spring wheat dwarfs. When grown in 18 hours of low light intensity per day and high temperature conditions winter wheat growth was vigorous with leaves measuring 40 to 50 inches in length. Under continuous high light intensity and lower temperatures, IBO 1535 headed at 10-14 inches and no leaf tips extended more than 16 inches from the surface of the pot. Under these conditions Zanda grew taller than IBO 1535 but showed no more than half the extension that it did under 18 hours of low light intensity and higher temperatures. IBO 1535 headed in 55 and 87 days, the latter being under cooler temperatures and continuous light of high intensity.

GENERAL DISCUSSION

In each experiment plants elongated and turned a pale yellow-green colour due to GA treatment. Applications of fertilizers overcame the yellowing but did not seem to correct the spindly stem and spike characteristic. Perhaps under more ideal light and temperature conditions, high soil fertility would produce stronger tillers and more seed.

Allan *et al.* (1) found that dwarf and semi-dwarf winter wheat varieties showed some growth response to GA but were shorter than normal. They found that normal plants responded more than dwarfs. Results of the present study conflict with those of Allan *et al.* because treated parents, Redman and Kenya Farmer, did not grow taller than the checks. Since the stage of growth at the time of GA application is important (4) then the repeated spraying throughout the development of the plants, which was used in Experiment III, would mask the effect of the one or two treatments which were applied at the correct stage. In the first experiment spraying at 46 days was much more successful than spraying at 75 days in the second experiment.

While it is not possible from the present study to trace the action of the GA it is possible to conclude that GA is useful in inducing heading and seed set in dwarf spring wheat.

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EFFECTS OF CLIPPING AND NITROGEN ON COMPETITION BETWEEN THREE PASTURE SPECIES[1]

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ABSTRACT

In three herbage mixtures comprised of different combinations of orchardgrass, *Dactylis glomerata* L., creeping red fescue, *Festuca rubra* L., and common white clover, *Trifolium repens* L., highest yields of the grasses were obtained when 1) harvesting was delayed until only 2 per cent of the incident light near mid-day penetrated to the base of the sward rather than at 10 per cent penetration, 2) a 2-inch rather han a 4-inch stubble was left after cutting, and 3) nitrogen fertilizer was applied. The same cutting treatments gave maximum yields of the associated clover without nitrogen fertilization, but when nitrogen was applied higher clover yields were obtained when cutting was at 10 per cent light penetration. Yields of the two grasses were approximately the same when grown with clover only, but in the two-grass association orchardgrass held the fescue to a low proportion in the mixture.

Results did not support the concept of maintaining a specified minimum leaf area index (LAI) for maximum yield of the herbage mixture.

INTRODUCTION

The existence of competition between herbage species growing in association implies that some requisite of growth is present in insufficient amount. If this limiting factor can be discovered and adjusted, increased yields may be possible. The objective of the experiment described herein was to study the effects of two factors, soil nitrogen and light intensity, as regulated by clipping, on the yields of orchardgrass, *Dactylis glomerata* L., creeping red fescue, *Festuca rubra* L., and common white clover, *Trifolium repens* L., grown in various associations.

In a previous experiment at Lethbridge (17) it was shown that when orchardgrass, creeping red fescue, and white clover were grown in association, the orchardgrass was so aggressive that it held the other two species to a low proportion in the mixture. It appears that orchardgrass was better able to compete for growth factors that were present in limiting amounts, and the suggestion was made that light and soil nitrogen might be the two most important limiting factors. The possible light deficiency was attributed to shading of the two lower-growing species by orchardgrass, and the nitrogen deficiency to the failure of the clover to fix enough atmospheric nitrogen.

One practical method of adjusting the light supply within the sward is through control of the height of cutting or grazing. Mitchell (8) expressed the view that, since pasture growth is basically a process of transferring light energy into plant tissue, it is important that all light be intercepted by photosynthetic tissue. He suggested that a basic inefficiency of many closely grazed pastures is the high proportion of the light energy that strikes bare soil. Brougham (2) attempted to test this hypothesis by measuring the rate of regrowth of ryegrass following defoliation to 5, 3,

[1]Contribution from the Forage Crops Section, Canada Agriculture Research Station, Lethbridge, Alberta, and the Farm Crops Department, Oregon State College, Corvallis, Oregon. Part of thesis presented by the senior author to the Graduate School, Oregon State College, Corvallis, Oregon, in partial fulfilment of the requirements for the Ph.D. degree.

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or 1 inches above ground level. The 5-inch stubble was sufficient to intercept 95 per cent of the incident light, and it was with this amount of stubble that the fastest rate of regrowth was observed over a 32-day period. Mitchell (10) gave even more convincing evidence for the validity of this hypothesis by showing that when white clover was either shaded 70 per cent, or left unshaded but defoliated 70 per cent, the amount of regrowth during the ensuing 15-day period was the same.

Despite evidence such as this, it is by no means generally agreed that less severe defoliation, as represented by greater stubble height, will lead to increased yields. Most experiments that have provided evidence in favour of greater stubble heights have been performed on spaced or potted plants, while experiments on dense pasture swards often have given the opposite results (7, 12, 14).

In dense pastures self-shading results in a sharp reduction in light intensities below the upper canopy of leaves (11). High intensities prevailing under sunny conditions may reach only a small portion of the leaves, and one would thus expect that the lower leaves would be less active photosynthetically. It has been suggested, in fact, that the lowest leaves might perform so little photosynthesis as to be parasitic on the rest of the plant (5). This could certainly be true of very young leaves, for Clendenning and Gorham (4) have shown that chloroplasts isolated from such leaves, although quite green, have no capacity for photosynthesis. Also, if these leaves were in deep shade they might remain parasitic for some time because, while chlorophyll content of leaves is low, photosynthesis proceeds slowly except at very high light intensities (6).

Low-growing species that are shaded almost continually by their taller associates are in a particularly disadvantageous position. Under reduced light not only does the plant as a whole have a lower capacity for photosynthesis but also the root-top ratio tends to decline (9, 13). Thus, the plant is less able to compete for soil nutrients and an accelerated loss of vigour occurs. Legumes may suffer a still further setback for it has been observed that when alfalfa is grown at very low light intensities the roots do not develop nodules (13).

When grass yields in a grass-clover mixture are increased through reducing the stubble height, it is difficult to say whether the increase results directly from the effects of clipping or indirectly from an improved soil nitrogen status due to better clover growth. The application of nitrogenous fertilizers to a mixed sward has not helped to distinguish between these effects because of the reduction in clover growth that results. Such reduction of clover usually has been attributed to increased shading by the grasses, or by weeds, although added nitrogen fertilizer may also reduce nodule formation by clover (1).

MATERIALS AND METHODS

A level, 1-acre site was selected on irrigated land at Lethbridge. The soil was classed as a Lethbridge fine sandy loam with good sub-surface drainage. It had been in alfalfa for 3 years prior to being ploughed in

August, 1956. The plots were broadcast-seeded in June, 1957, and good stands were obtained. Mixtures seeded and rates of seeding, in pounds per acre, were as follows:

1) orchardgrass 8, white clover 3;
2) creeping red fescue 10, white clover 3; and
3) orchardgrass 4, creeping red fescue 5, white clover 3.

The plots were 30 x 54 feet and were located at random within each of three replicates.

The main blocks of each mixture were divided into eight sub-plots, 6.75 x 30 feet, and treatments were assigned at random to them. These treatments, applied in all combinations, were as follows:

Treatment No.	Light level when cut	Height of stubble	Nitrogen fertilizer used
	percent	inches	pounds
1	10	2	None
2	2	2	None
3	10	4	None
4	2	4	None
5	10	2	100
6	2	2	100
7	10	4	100
8	2	4	100

Ammonium nitrate fertilizer (33.5-0-0) was broadcast annually in three 100-pound applications at 6-week intervals beginning May 1. The light percentage at the time of cutting refers to light penetration through the sward to a point one-half inch above the soil surface. The levels were 10 per cent and 2 per cent of the incident light falling on a horizontal plane in the open. Light intensities were measured by use of a Gossen, Tri-Lux, foot-candle meter, Model C. In this instrument the photoelectric cell is on a 3-foot extension cord from the microgalvanometer. The spectral sensitivity is approximately that of the human eye.

Light measurements were made between 11.00 a.m. and noon and between 1.00 and 2.00 p.m. when possible, although it was necessary to deviate from this schedule occasionally when cloud cover interfered.

Yields were determined by weighing the fresh herbage from a 34-inch mower strip and taking a sample of approximately 600 grams for dry matter determination. The remaining herbage of the plot was then mowed to the same stubble height and discarded. Each plot was cut as it reached its prescribed light level without regard to the same treatments in other replications.

Botanical composition of the harvested herbage was determined for each plot at each cutting by visual estimation of the sample drawn for dry matter determination. The percentages of each species were used to calculate individual species yields.

Leaf area index (LAI), the leaf area per unit area of soil surface (16), was calculated for each mixture at various stages of growth. The LAI was estimated for each species in each 1-inch horizontal segment of the sward. This was done on a known area of sod that was suspended upside down on a ring stand in the laboratory. The leaves of each species in the sod were clipped into successive 1-inch segments and weighed separately. The leaf area was determined by multiplying the weight by a factor that related weight to area. Only laminae were considered as leaves. The petioles of the clover and the sheaths of the grasses were discarded. This is in accordance with the definitions of Stapledon (15) and Brougham (2).

Plots were irrigated regularly to maintain good soil moisture conditions. Applications of 200 pounds of triple superphosphate, 0-46-0, were made in the spring of 1957 and 1958 to obviate any possible phosphorus deficiency.

RESULTS

The necessity of mowing to control weeds early in the season caused some delay in beginning the treatments in 1958. The first regular harvests were made in late June. Despite the short season the results for 1958 were essentially the same as those for 1959 and will not be discussed except where necessary to point out differences that occurred between the 2 years.

The first harvest of 1959 was on May 22, and a final harvest of all plots was made on September 23. The number of harvests of any one plot ranged from 3 to 12, depending on the treatment. The most frequently harvested plots were those fertilized with nitrogen and cut back to a 4-inch stubble when light penetration was 10 per cent of full daylight. The least frequently harvested were the no-nitrogen, 2-inch stubble, 2 per cent light penetration plots.

Height of the herbage at the time of harvest varied over the season. The same light interception occurred with a shorter growth as the season progressed, owing mostly to the lower elevation of the sun later in the season. By the end of August the 4-inch stubble was sufficient to reduce the light at ground level below 10 per cent of full daylight. A departure from the prescribed cutting treatment was necessary, so the procedure adopted after the end of August was to postpone cutting until a final harvest of all plots was made on September 23.

The first spring harvest of the 2 per cent light treatment plots was late because light penetration through the foliage remained high. By mid-June the orchardgrass had begun to flower and had reached a height in excess of 36 inches on some plots, but still the light at ground level had fallen to only about 5 per cent of full light. It seemed that a loss in yield would result from a further delay in cutting, so all of the 2 per cent light plots were harvested on June 16. Ten per cent light penetration occurred with approximately 13 inches of growth at that time of year. As an average for the season the height of the grasses when the leaves were held erect was 11 to 13 inches at 10 per cent light penetration and 15 to 18 inches at 2 per cent penetration. The height of the fescue tended toward the lower figure and that of the orchardgrass toward the higher. When the herbage

TABLE 1.—EFFECTS OF MAIN TREATMENTS ON THE YIELDS OF EACH COMPONENT OF THREE HERBAGE MIXTURES IN 1959

	Light penetration		Stubble height		Pounds per acre nitrogen	
	10%	2%	2″	4″	0	100
	Pounds of dry matter per acre					
Mixture 1						
Orchard	2668	4290*	3816	3142*	2710	4248*
Clover	1359	1168	1773	754*	1698	829*
Total[1]	4096	5525*	5684	3937*	4480	5142*
Mixture 2						
Fescue	2635	3900*	3912	2623*	2705	3830*
Clover	1862	2086	2786	1161*	2293	1655*
Total[1]	4842	6397*	7220	4019*	5317	5922*
Mixture 3						
Orchard	2877	3971*	3867	2981*	2500	4348*
Fescue	235	270	275	230	210	295*
Clover	1532	1368	1911	990*	1919	982*
Total[1]	4734	5702*	6172	4264*	4706	5730*

[1]Includes weeds, not listed here
*Difference between contrasting treatments significant at the 1 per cent level

was measured as it stood, without holding the leaves erect, corresponding heights were 7 to 7.5 and 8 to 10 inches for 10 per cent and 2 per cent light penetration, respectively.

Dry matter yields of each component of the mixtures are grouped by treatment means in Table 1. When cutting was delayed until only 2 per cent of full daylight penetrated the sward, yields of the grasses increased but clover yields were not affected. With only one exception, leaving a 2-inch stubble resulted in higher yields of both grasses and clover than leaving a 4-inch stubble. The effect of the nitrogen treatment was significant in all cases, the yields of the grasses being increased and those of the clover decreased. A significant interaction occurred between light and stubble height in Mixture 1, due to the fact that the detrimental effect of the 4-inch stubble was partially offset by allowing growth to reach the 2 per cent light penetration stage before cutting. The interaction of light and nitrogen was significant for some components, due to a greater effect of nitrogen under the 2 per cent light treatment and of the 2 per cent light treatment when nitrogen was applied. The effect of both these treatments was to increase the yield of the grasses and decrease the yield of clover. Significant stubble x nitrogen interactions resulted from the greater effect of nitrogen fertilizer under a 2-inch stubble than under a 4-inch. In addition, the application of nitrogen increased the effect of stubble height on grass yields and decreased the effect on clover yields.

An indication of the seasonal distribution of growth was obtained by dividing the season into four 33-day periods and summing the yields within each period. Cumulative yields for the season, beginning at zero on May 14, were plotted on co-ordinate paper, and the yield at the end of each 33-day period was read on the y-axis. Data for each component of

TABLE 2.—YIELDS OF INDIVIDUAL COMPONENTS OF TWO MIXTURES UNDER EIGHT MANAGEMENT TREATMENTS DURING EACH OF FOUR 33-DAY PERIODS FROM MAY 14 TO SEPTEMBER 23, 1959

Treatment	Pounds of dry matter per acre				
	Period 1	Period 2	Period 3	Period 4	Total
Orchardgrass in Mixture 1					
10%;2";N/0	1118	761	567	124	2570
2%;2";N/0	1952	752	438	274	3416
10%;4";N/0	527	726	328	52	1633
2%;4";N/0	2153	517	418	133	3221
10%;2";N/100	1630	1217	598	318	3763
2%;2";N/100	3103	1337	732	347	5519
10%;4";N/100	886	1336	375	111	2708
2%;4";N/100	2941	1110	654	299	5004
Clover in Mixture 1					
10%;2";N/0	1228	842	197	33	2300
2%;2";N/0	1047	977	329	159	2512
10%;4";N/0	217	686	40	9	953
2%;4";N/0	321	544	101	28	994
10%;2";N/100	767	689	119	25	1600
2%;2";N/100	260	208	142	36	646
10%;4";N/100	41	517	21	4	583
2%;4";N/100	156	233	77	19	485
Fescue in Mixture 2					
10%;2";N/0	1012	836	662	250	2760
2%;2";N/0	1743	950	642	220	3555
10%;4";N/0	716	611	403	84	1814
2%;4";N/0	1016	802	555	273	2691
10%;2";N/100	1389	1098	1180	419	4086
2%;2";N/100	2463	1412	912	462	5249
10%;4";N/100	400	833	495	155	1883
2%;4";N/100	1813	1137	853	302	4105
Clover in Mixture 2					
10%;2";N/0	1357	1175	365	121	3018
2%;2";N/0	1542	1441	414	87	3484
10%;4";N/0	240	973	145	11	1369
2%;4";N/0	397	732	147	24	1300
10%;2";N/100	1020	717	378	66	2181
2%;2";N/100	1050	953	365	88	2456
10%;4";N/100	72	715	84	8	879
2%;4";N/100	370	574	123	29	1096

Mixture 1 and 2 are recorded in Table 2. The most striking result is the low yield of the 10 per cent light, 4-inch stubble treatment during period 1. In most cases yields under this treatment compared favourably with those under other treatments during subsequent periods, but the loss that occurred during these first 33 days, as much as 3,261 pounds of dry matter per acre for the combined fescue and clover yields in Mixture 2, could not be made up. The other conspicuous aspect of Table 2 is the decline in yields as the season progressed. Forty-eight and forty per cent of the average yield

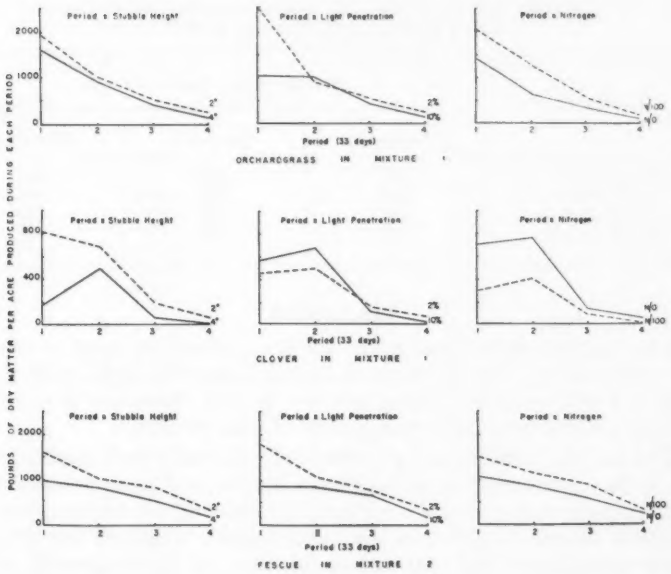

FIGURE 1. Effects of various treatments on yield as the season progressed.

of Mixtures 1 and 2, respectively, was produced during the first 33 days, and eighty and seventy-five per cent was produced during the first half of the season.

The effects of the treatments on the distribution of seasonal yields were determined by analysis of variance of the yields for each period and treatment. The trends are illustrated in Figure 1. Most of the significant interactions occurred because differences due to treatments were greater during period 1 than they were later in the season. After the first period the effects of the treatments apparently were minimized by the large influence of factors other than those used as variables.

The question arose as to whether the amount of material left unharvested in the 4-inch stubble treatment might account for the yield deficit of the 4-inch as compared with the 2-inch stubble. To provide information on this point all 4-inch stubble treatments were reharvested to a 2-inch stubble at the conclusion of the regular cutting schedule on September 23. Yield data are recorded in Table 3.

Results of some of the LAI (leaf area index) determinations are recorded in Table 4. The data show the distribution of the leaves of each species in the canopy. It is evident that a considerable proportion of the leaf area of all species, but clover particularly, was below the 4-inch level. No leaf was present below the 2-inch level.

TABLE 3.—YIELDS OBTAINED BY MOWING 4-INCH STUBBLES TO A 2-INCH STUBBLE ON SEPTEMBER 23, 1959

Treatment	Pounds of dry matter per acre		
	Mixture 1	Mixture 2	Mixture 3
10%;4";N/0	276	628	318
2%;4";N/0	329	808	491
10%;4";N/100	368	693	390
2%;4";N/100	672	1100	437

DISCUSSION

The fact that light interception was the criterion of stage of development does not deny the influence of factors other than light, such as temperature, humidity, and clipping *per se*. In this discussion it is assumed that light was the most influential quality of solar radiation.

The marked yield advantage associated with the 2-inch stubble is contrary to the hypothesis that with a higher stubble, and more photosynthetic tissue, the plant is able to make more rapid regrowth. It might appear that the greater amount of clover in the 2-inch stubble treatment improved the soil nitrogen supply and was responsible for the better growth of the grasses, but this possibility seems unlikely for several reasons. First, grass yields were higher from the 2-inch stubble treatment in 1958 before the clover had a chance to exert much influence, and the same was true during the first 33-day period of 1959. In the second place, if the nitrogen contributed by the clover was responsible for the improved growth of the grass, one would expect the most response by the grass to nitrogen under the 4-inch stubble, which is opposite to the results actually obtained.

The hypothesis of more photosynthetic tissue leading to a faster growth rate cannot be discarded. It is supported here by the fact that the 2 per cent light penetration treatment led to higher yields than the 10 per cent treatment under comparable stubble and nitrogen treatments. The difference between the effect of the 2 per cent and the 10 per cent light treatments may have been in part a manifestation of differences in intervals between cutting. The 10 per cent light, 2-inch stubble treatment was harvested an average of six times during the season, and the 2 per cent light, 4-inch stubble treatment an average of four times. The yields of orchardgrass in Mixture 1 were greater with less frequent clipping, but orchardgrass in Mixture 3 and fescue in both mixtures did not appear to be influenced directly by clipping frequency.

None of these observations supports a reason for the grasses yielding more under a 2-inch than a 4-inch stubble. The fact that more of the plant is harvested accounts for some of the difference, although by no means all (Tables 1 and 3). A high light intensity close to the base of the plant after cutting might be important. When the sward is cut, the older, more photosynthetically efficient, leaf tissue is removed and the younger, less efficient, tissue is left. As suggested earlier, this younger tissue is efficient at photo-

TABLE 4.—LAI OF EACH SPECIES IN SUCCESSIVE 1-INCH SEGMENTS OF THE SWARD OF AN ORCHARDGRASS-FESCUE-CLOVER MIXTURE AT FOUR SAMPLING LOCATIONS

Height interval (inches)	Sample 1			Sample 2		
	Orch.	Fesc.	Clov.	Orch.	Fesc.	Clov.
20-19	0.013			0.006		
19-18	0.013			0.006		
18-17	0.019			0.013		
17-16	0.025			0.013		
16-15	0.069			0.019		
15-14	0.132			0.032		
14-13	0.139			0.057	0.003	
13-12	0.164			0.063	0.003	
12-11	0.233			0.101	0.003	
11-10	0.303	0.003		0.151	0.069	
10-9	0.366	0.003		0.183	0.069	
9-8	0.448	0.003		0.265	0.017	
8-7	0.511	0.007		0.258	0.021	
7-6	0.574	0.017	0.346	0.347	0.021	0.346
6-5	0.744	0.021	0.676	0.473	0.038	0.626
5-4	0.725	0.024	0.338	0.498	0.055	0.585
4-3	0.662	0.017	0.437	0.504	0.107	0.371
3-2	0.365	0.010	0.058	0.158	0.076	0.066
2-1	0.000	0.000	0.000	0.000	0.000	0.000
1-0	0.000	0.000	0.000	0.000	0.000	0.000
Total	5.517	0.122	1.855	3.147	0.416	1.994
LAI for mixture		7.494			5.551	
		Sample 3			Sample 4	
12-11	0.006					
11-10	0.063	0.003				
10-9	0.082	0.007				
9-8	0.126	0.007		0.038		
8-7	0.183	0.031	0.074	0.044		
7-6	0.252	0.052	0.321	0.139	0.007	
6-5	0.328	0.052	0.338	0.252	0.014	0.025
5-4	0.322	0.045	0.247	0.700	0.038	0.338
4-3	0.523	0.086	0.190	1.128	0.024	0.379
3-2	0.637	0.107	0.255	1.564	0.031	0.445
2-1	0.000	0.000	0.000	0.000	0.000	0.000
1-0	0.000	0.000	0.000	0.000	0.000	0.000
Total	2.522	0.390	1.425	3.865	0.114	1.187
LAI for mixture		4.337			5.166	

Note: The height of each species is indicated by the maximum height interval in which it occurred.

synthesis only at very high light intensities. These intensities would be available to all of the above-ground tissue under the 2-inch cutting but, because of self-shading, to only a portion of the tissue under a 4-inch cutting. The lowermost tissue of the 4-inch stubble might be 'parasitic' and slow down the regrowth of the plant as a whole. Support for this suggestion of the importance of timeliness of high light availability is given by the clover reaction. Clover growth was reduced by the higher stubble treatment but usually was not affected by the height that the grasses reached prior to cutting. In most cases clover yields were approximately the same under the 2 per cent and 10 per cent light treatments (Table 1).

If a high light intensity near the base of the plant is important it might help to explain why spaced and potted plants usually yield more with higher stubbles. Light is available to spaced and potted plants around the perimeter as well as from above and so is a lesser variable than it is in a dense sward. Spaced plants with a high stubble can take advantage of the greater amount of photosynthetic tissue they process.

The competitive advantage of orchardgrass over fescue was not reduced by any of the treatments, possibly because the species were affected in the same manner by the treatments. A somewhat different situation existed in the orchardgrass-clover relationship. Where no nitrogen was applied both species tended to be favoured by the same treatments but, in addition, clover had the advantage of an independent source of nitrogen. Therefore, clover yields were high under the 2 per cent light, 2-inch stubble, no-nitrogen treatment. When nitrogen fertilizer was added, and this advantage nullified, clover was a less successful competitor.

The failure of the different management treatments to eliminate the dominance of orchardgrass over fescue may be attributable to the fact that the proportions of the two species were established soon after seeding. This possibility had been considered from the beginning, and low seeding rates of equal numbers of seeds of each species were used in an attempt to avoid it. The original stand was indeed sparse; nevertheless, the proportions of the two grasses had been established early in 1958.

Within the season, also, the major influences of the treatments were manifest early. In one case, leaving 4 inches rather than 2 inches of stubble resulted in a yield difference of more than 1 ton per acre within the first 33 days (Table 2). The deficit was not made up; in fact, it increased to 2 tons by the end of the season. This point is important in pasture management. When pastures are divided to permit rotational grazing, a common practice in spring is to move the animals quickly from one field to another, leaving a high stubble after grazing. The better solution, in view of these results, would be to graze to a shorter stubble height and conserve as hay or silage any excess that cannot be grazed immediately. The beneficial effect of the 2 per cent light treatment on the grasses occurred almost entirely within the first 33 days. Here again a suggested principle is often contravened in pasture management, for it is common practice to begin grazing in spring when the grass is short, in order to prevent a surplus from occurring. The method may be effective, but higher yields could be expected by delaying grazing until growth is well advanced. The most pronounced effect of nitrogen was reduction of the yield of clover, and it occurred mainly during the first half of the season.

The importance of the LAI measurements is diminished with the realization that the lower stubble height was the more productive. Certainly, the concept that maintenance of a specified minimum LAI will lead to a maximum rate of growth was not supported. With a 2-inch stubble the LAI was zero; yet good regrowth occurred. The data on the relationship between LAI and light interception agree fairly well with those presented by Brougham (3) for a part of the season. With the sun at its maximum elevation above the horizon the mixed herbage intercepted ap-

proximately 90 and 98 per cent of the incident light with LAI of about 4 and 7, respectively. The relationship was determined directly by the elevation of the sun, however, for shortly after mid-August, when the sun was lower in the sky, 90 per cent of the incident light was intercepted by herbage with an LAI of little more than 1. Here, of course, the sheaths, which were not included in the leaf area measurements, were responsible for a considerable proportion of the light interception.

Elevation of the sun may be of fundamental importance in explaining the difference between these results and those obtained by Brougham. Brougham's experiments were conducted at a latitude of 40° 30′ S., where the noon elevation of the sun exceeds the maximum reached at Lethbridge (50° N.) for approximately 2 months of the year. The differences would favour the maintenance of a higher LAI for grasses under New Zealand conditions.

Another consideration is the species employed. Small differences between species in orientation of the leaves might alter light interception considerably.

CONCLUSIONS

1) The beneficial effect of the 2-inch over the 4-inch stubble was real. It may have resulted from the need of a high light intensity near the base of the plants for the initiation of regrowth or from the removal of old, non-functional plant material that shaded the younger, functional leaves. These points merit further investigation.

2) Shading by tall growing orchardgrass was not an important factor in reducing the growth of associated clover and fescue.

3) Clipping practices that permitted high light penetration into the sward and the maintenance of a high level of soil nitrogen did little to reduce the dominance of orchardgrass over creeping red fescue growing in association.

4) The concept of maintenance of a specified minimum leaf area for a maximum rate of regrowth was not supported.

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PHENOLOGY OF SEVERAL PLANT SPECIES AT OTTAWA, ONTARIO, AND AN EXAMINATION OF THE INFLUENCE OF AIR TEMPERATURES[1]

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ABSTRACT

The apparent base temperature, below which plants do not develop towards flowering, was estimated for 10 of the early flowering woody species by means of a modified linear regression analysis. The relationship between average spring temperatures and developmental rates was examined for the same 10 species. The rate of development towards flowering in *Acer saccharinum*, *A. saccharum*, *Populus tremuloides*, *P. grandidentata* and *Ulmus americana* was more closely related to average maximum temperatures while in *Acer rubrum* and *Betula papyrifera* it was more closely related to minimum temperatures. *Alnus rugosa*, *Acer negundo* and *Prunus pensylvanica* were not markedly influenced by average spring temperatures. The first dates of flowering are also presented for 36 plant species that were observed during the period 1936-1960 at Ottawa, Ont. The marker plants include 18 trees, 6 shrubs, 6 grasses and 6 other herbaceous and weedy species.

INTRODUCTION

Phenological responses of plants have been the interest of many workers in forestry and agriculture. Not only is this information of importance to those who investigate the growth and control of plants but it has value for the entomologist dealing with insect problems on plants and for the allergist studying pollen induced allergies.

Minshall (6) briefly discussed the relationship between the recorded mean monthly temperature and the first flowering of 4 trees at Ottawa for the period 1936 to 1945. More recently Budd and Campbell (2) and Moss (7) have reported the results of phenological work at Swift Current, Saskatchewan, and Edmonton, Alberta, respectively. Nuttonson's (8) research on the relationship between weather and wheat phenology is a classic. Lindsey and Newman (5) used a temperature summation technique to explain variation in flowering of species in Indiana. It was not based on means, but on daily maximum and minimum temperatures on the assumption that growth was linear. This method excels use of the mean in summation for days when rising or falling temperature crosses the threshold temperature level of the plant. It was not applicable to the data of Moss (7) at Edmonton, Alberta, who used a simpler summation method utilizing maximum temperature, to explain variation in flowering of aspen, pin cherry and choke cherry. Ahlgren (1) sought to relate daily maximum temperature to beginning of growth activity in tree species. Some trees began growing even though daily minimum temperatures were below freezing. The present paper is an attempt to relate first dates of anthesis of 10 early flowering woody plants at Ottawa with air temperature data for the period 1936-1960. Phenological data on 26 other plant species are also presented.

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[1]Contribution No. 114 from the Plant Research Institute, Research Branch, Canada Department of Agriculture, Ottawa, Ont.

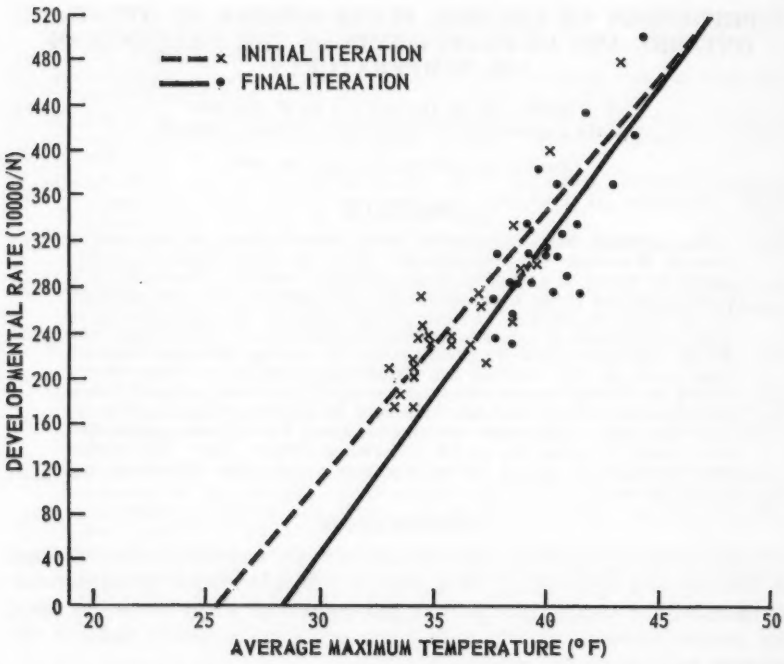

FIGURE 1. Regression lines and bivariate distributions showing the relationship between average maximum temperature and developmental rate in *Acer saccharinum*. (N = number of days above threshold temperature from March 1 to flowering date.)

METHODS

The study reported herein was carried out in the vicinity of the Dominion Arboretum located at the Central Experimental Farm, Ottawa. For phenological purposes, the date of flowering was the first opening of the stamens on a designated plant or group of plants. Many of the marker plants originally selected by Minshall (6) were used and his procedure was followed for the selection of additional plants. In the case of the few annual species that were observed, a small group for each species was selected from the same locality and habitat each year. The plant names herein are those used by Fernald (3). A summary of the first flowering dates for the 36 species is presented in Table 3. The weather records were obtained from the Agrometeorological Section, Plant Research Intsitute, Central Experimental Farm, Ottawa.

The species under investigation were grouped into three general categories with regard to first flowering date: Group 1 (March and April), Group 2 (May and June) and Group 3 (July, August and September). A preliminary study of the data also showed that the variation in flowering date of species in Group 1 and some of Group 2 could be explained largely

on the basis of temperature alone. None of the species in Group 3 yielded satisfactorily to this type of analysis. This paper, therefore, will deal only with the results of temperature-flowering date relationships of 10 species in Groups 1 and 2.

In the present paper the apparent base temperature of the species, below which the plant does not develop towards flowering, was estimated by a repeated regression analysis. This technique was used to eliminate non-development days when temperatures were below the apparent threshold value. March 1 was chosen as the starting time for the calculations because very few days occur before that date on which the temperatures are high enough for growth or development.

The number of effective days, between the starting date and the flowering date, with the temperature above the apparent threshold was designated by the symbol N. The average rate of development on effective days during this period was computed as 10,000/N. The objectives of the regression analyses were to estimate: 1) the apparent threshold temperature and 2) the number of developmental days with temperature above the apparent threshold.

The initial threshold value was set very low so that all days were included in the calculations. Thus, N was equal to the total number of days between March 1 and the flowering date. The apparent threshold was then estimated by solving for the constants in the regression equation

$$10{,}000/N = a + bT$$

where T is either the average minimum or maximum temperature during the developmental period prior to flowering. This equation can be written in the form

$$10{,}000/N = b\left(T + \frac{a}{b}\right)$$

or

$$10{,}000/N = b\left(T - \left(-\frac{a}{b}\right)\right)$$

The term $-a/b$ is the apparent threshold temperature. Graphically it is the point where the regression line crosses the temperature axis (see Figures 1 and 2).

The initial threshold value was replaced by this first estimate; all daily temperatures were re-examined and those below the threshold were rejected. From the remaining values a new maximum (or minimum) for the development period was calculated as well as a new value for N. The regression analysis was repeated and a second estimate of the threshold temperature determined. This procedure was repeated until all remaining temperatures were above the last apparent threshold calculated. This final value was then accepted as the best estimate of the apparent threshold temperature.

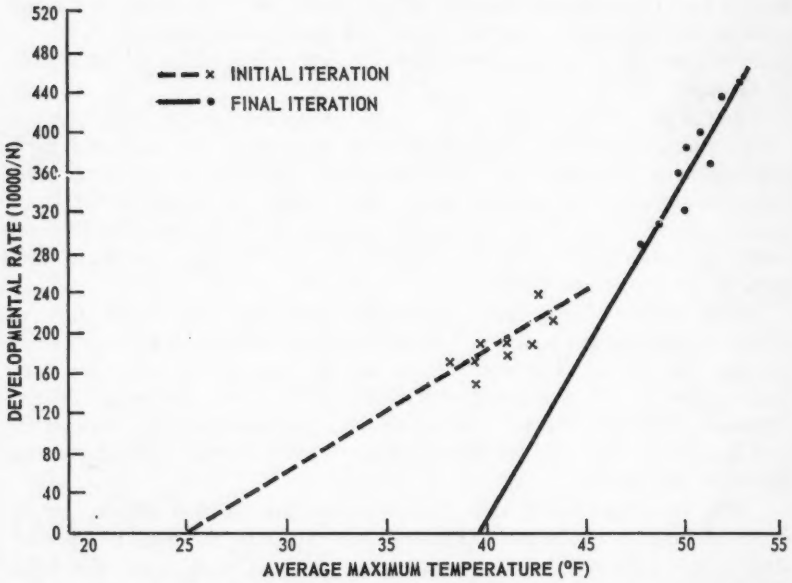

FIGURE 2. Regression lines and bivariate distributions showing the relationship between maximum temperature and developmental rate in *Populus grandidentata*. (N = number of days above threshold temperature from March 1 to flowering date.)

An example of the analysis of the influence of average maximum temperature on developmental rate for *Populus tremuloides* is presented in Table 1. On the first iteration a very low threshold was used so that no days were deleted. The calculated threshold was inserted for the second iteration and all daily temperatures below 23°F. were subsequently deleted. A new rate (10,000/N) was computed for each year and another regression analysis was performed. The regression analyses were repeated 4 times and provided a final estimated threshold maximum of 27.9°F. for *P. tremuloides*.

Daily temperature records were available on punch-cards and the analysis was carried out on an electronic computer.

RESULTS

The apparent threshold temperatures for 10 of the early spring flowering species are presented in Table 2. The coefficient of determination is the square of the coefficient of correlation and represents the proportion of the variation in developmental rate that was accounted for by variation in average temperature. For example, variations in average maximum temperature account for approximately 88 per cent of the changes of flowering date of *Populus grandidentata*.

DISCUSSION

It was hoped that this study could be conducted by examining only maximum daily temperatures and their relation to flowering. However,

it was found that in some cases minimum temperatures were much more closely related to developmental rate. Went (9) has shown for several plant species that variabilities in temperature play an important part in the initiating of flowering and plant development. Accordingly, the analysis of both sets of temperature records are presented for purposes of comparison. In some cases the initial run resulted in an estimate of a threshold temperature below any of the temperatures encountered subsequent to March 1. No further iterations were attempted with these species. The magnitude of the coefficient of determination indicated, to a large extent, the precision with which the apparent threshold was estimated.

It should be emphasized that the apparent threshold temperatures were computed on the assumption that a linear relationship existed between temperature and developmental rate. The possibility that the linear relationship was not applicable in the region of the apparent threshold temperatures must be recognized. However, the technique provided realistic threshold temperatures for several species.

As shown in Table 3, *Alnus rugosa* was the first species to flower in the early spring. The coefficient of determination as shown in Table 2 for

TABLE 1.—NUMBER OF DEVELOPMENTAL DAYS CONSIDERED AND STATISTICS CALCULATED DURING EACH ITERATION OF A STUDY OF THE RELATIONSHIPS BETWEEN AVERAGE MAXIMUM TEMPERATURE AND FLOWERING DEVELOPMENTAL RATE OF *Populus tremuloides*.

Year	Number of developmental days included in each iteration			
	First	Second	Third	Fourth
41	45	39	35	33
42	48	48	46	46
43	62	55	52	51
44	57	48	46	46
45	29	27	27	27
46	29	28	28	28
47	57	56	54	53
48	49	43	41	41
49	44	38	33	33
50	58	49	45	44
51	51	51	46	45
52	49	45	42	42
53	36	31	30	30
54	50	46	41	41
55	46	39	36	35
56	58	51	49	49
57	52	50	48	48
58	42	42	41	41
59	51	45	44	43
60	53	44	42	39
Estm'd. threshold (°F.)	23.6	26.8	27.5	27.9
Regression coef.	13.7	16.3	16.9	17.4
R^2	0.64	0.64	0.59	0.58
Average maximum	39.3	41.4	42.3	42.5
Average rate $\left(\frac{10000}{N}\right)$	216.	238.	251.	255.

TABLE 2.—APPARENT THRESHOLD TEMPERATURES, REGRESSION COEFFICIENTS, AND COEFFICIENTS OF DETERMINATION RESULTING FROM AN EXAMINATION OF THE RELATIONSHIPS BETWEEN AIR TEMPERATURE AND DEVELOPMENTAL RATES OF 10 WOODY SPECIES

	Apparent threshold (°F.)		Regression coefficient		Coefficient of determination (R^2)		R^2 required for significance		No. of years	No. of iterations	
	Max.	Min.	Max.	Min.	Max.	Min.	.05	.01		Max.	Min.
Alnus rugosa	8.7	−13.4	10.2	8.0	0.31	0.44	0.44	0.64	9	1	1
Acer saccharinum	28.4	1.4	27.3	13.2	0.62	0.26	0.16	0.27	24	4	4
Populus tremuloides	27.9	−9.2	17.4	6.7	0.58	0.17	0.20	0.31	20	4	2
Populus grandidentata	39.6	9.3	34.3	11.6	0.88	0.64	0.44	0.64	9	9	5
Ulmus americana	30.7	−5.1	16.0	6.3	0.54	0.17	0.16	0.27	24	4	3
Acer rubrum	24.7	6.5	10.3	9.2	0.51	0.79	0.44	0.64	9	3	3
Betula papyrifera	4.2	−2.0	4.1	5.6	0.29	0.52	0.44	0.64	9	1	4
Acer negundo	16.3	−40.7	6.4	2.6	0.31	0.10	0.20	0.31	20	2	1
Acer saccharum	28.6	−8.4	8.9	4.2	0.50	0.17	0.20	0.31	20	4	2
Prunus pensylvanica	−4.4	−18.1	2.7	2.9	0.20	0.26	0.21	0.33	19	1	2

TABLE 3.—FIRST FLOWERING DATES FOR 36 PLANT SPECIES AT OTTAWA, ONTARIO

Species	Years of observation	First flowering date			Range of first flowering dates in days
		Mean date	Earliest date and year	Latest date and year	
			Group No. 1		
Alnus rugosa	9 (1952-1960)	April 7	March 29, 1958	April 14, 1959	16
Acer saccharinum	25 (1936-1960)	April 11	March 21, 1946	April 26, 1940	36
Populus tremuloides	20 (1941-1960)	April 16	March 29, 1946	April 27, 1956	29
Corylus cornuta	8 (1953-1960)	April 16	April 4, 1953	April 23, 1956	19
Populus grandidentata	9 (1952-1960)	April 24	April 11, 1953	May 5, 1956	24
Ulmus americana	25 (1936-1960)	April 26	April 7, 1946	May 7, 1943	30
Acer rubrum	9 (1952-1960)	April 26	April 18, 1958	May 8, 1956	20
Poa annua	9 (1952-1960)	April 26	April 13, 1955	May 1, 1956	18
			Group No. 2		
Betula papyrifera	9 (1952-1960)	May 2	April 23, 1958	May 14, 1956	21
Acer negundo	20 (1941-1960)	May 5	April 9, 1945	May 12, 1956	33
**Acer saccharum*	25 (1936-1960)	May 9	April 14, 1946	May 21, 1956	37

**Acer saccharum* did not flower in the years 1946, 1951, 1957 and 1958.

(*Table 3 continued on pp. 650 and 651*)

TABLE 3.—*continued*

Species	Years of observation	First flowering date			Range of first flowering dates in days
		Mean date	Earliest date and year	Latest date and year	
			Group No. 2		
Celtis occidentalis	7 (1952-1958)	May 12	May 1, 1951	May 30, 1956	29
			1942		
Prunus pensylvanica	19 (1942-1960)	May 13	May 2, 1945	May 28, 1956	26
Barbarea vulgaris	9 (1952-1960)	May 14	May 5, 1955	May 22, 1956	17
Alopecurus pratensis	9 (1952-1960)	May 14	May 8, 1954	May 28, 1956	20
Fagus grandiflora	8 (1953-1960)	May 15	May 11, 1955	May 28, 1956	17
Smilacina stellata	19 (1942-1960)	May 20	May 8, 1942	May 31, 1956	23
Quercus macrocarpa	9 (1952-1960)	May 22	May 16, 1953	June 4, 1956	19
			1942		
Pinus sylvestris	25 (1936-1960)	May 27	May 18, 1955	June 10, 1957	23
			1953		
Poa pratensis	9 (1952-1960)	May 28	May 19, 1957	June 12, 1956	24
Rumex acetosella	8 (1952-1959)	June 3	May 26, 1955	June 12, 1956	17
				1947	
Anemone canadensis	18 (1942-1959)	June 3	May 26, 1951	June 12, 1956	17
Juglans nigra	9 (1952-1960)	June 7	May 29, 1960	June 14, 1956	16
				1956	
Dactylis glomerata	9 (1952-1960)	June 9	May 29, 1959	June 17, 1958	19
				1947	
Carya cordiformis	16 (1945-1960)	June 12	June 6, 1955	June 23, 1956	13

TABLE 3.—*continued*

Species	Years of observation	First flowering date			Range of first flowering dates in days
		Mean date	Earliest date and year	Latest date and year	
			Group No. 2		
				1950	
Sambucus nigra	11 (1950-1960)	June 16	June 8, 1955	June 19, 1958	11
Bromus inermis	19 (1942-1960)	June 19	June 10, 1955	June 28, 1947	18
Phleum pratense	19 (1942-1960)	June 25	June 18, 1955	June 30, 1956	12
			1949		
Rhus typhina	14 (1947-1960)	June 26	June 17, 1955	July 9, 1956	22
			Group No. 3		
				1956	
Catalpa ovata	17 (1944-1960)	July 2	June 17, 1955	July 17, 1958	30
Tilia americana	19 (1941-1960)	July 5	June 23, 1959	July 18, 1947	25
Ambrosia trifda	9 (1952-1960)	July 13	July 4, 1955	July 26, 1958	22
Cephalanthus occidentalis	15 (1946-1960)	July 18	July 10, 1955	July 30, 1956	20
			1949		
Cassia hebecarpa	14 (1947-1960)	Aug. 3	July 27, 1953	Aug. 13, 1952	17
Ambrosia artemisiifolia	8 (1953-1960)	Aug. 7	Aug. 1, 1957	Aug. 13, 1954	12
Hamamelis virginiana	17 (1944-1960)	Sept. 19	Sept. 4, 1953	Oct. 6, 1947	32

this species was very low for both maximum and minimum temperatures. An examination of the staminate catkins in October revealed that meiosis had already occurred and the pollen appeared to be well developed and mature. This suggests that the flowers were quite dormant at the onset of winter but by the time the first few warm days arrived in early spring this dormancy had been broken and anthesis followed immediately. Higgins and Arisumi (4) observed with several species of *Ulmus*, including *Ulmus americana*, that dates of first floral differentiation in buds occurred during the summer previous to the next year of flowering. The megaspore and microspore mother cells had been differentiated by October but had passed through the winter in this stage without further enlargement. Apparently temperature over a longer period in the spring plays a more important part in the flowering of *Ulmus americana* than in the flowering of *Alnus rugosa*.

This technique appears to work well for some species (e.g. *Populus grandidentata*) implying that average early spring temperatures are an important factor in governing the time of flowering in certain woody species. However, flowering in some species was not closely related to temperature. In these cases a more comprehensive study is needed to examine other factors in relation to flowering.

This study was limited to an examination of the relationship between temperatures and date of flowering. Although other factors probably contribute to the yearly variation in flowering dates, temperature appears to be one of the main factors, especially for those species flowering early in the spring.

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AN EVALUATION OF VARIETAL CHARACTERISTICS OF ORCHARDGRASS (*DACTYLIS GLOMERATA* L.) SUBJECTED TO A SILAGE-PASTURE TYPE OF UTILIZATION

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ABSTRACT

A study was conducted over 3 harvest years to evaluate the importance of maturity classification, tillering, and fall dormancy in relation to yield of dry matter, per cent protein and yield of protein, and regional adaptation of 40 orchardgrass varieties subjected to a silage-pasture type of utilization. Using head emergence (silage stage) as the criterion, five maturity classes were established: *very early*, *early*, *medium*, *late*, *very late*. Early maturity was positively associated with total yield, but there was no relation between maturity and per cent protein, tillering, or fall dormancy. Total yield of dry matter was the main determinant of protein yield. Tillering was not associated with yield of dry matter or per cent protein but was associated with fall greenness. Fall dormancy was positively associated with prostrate fall growth habit. Individual varieties displaying desirable agronomic characteristics were found within every maturity class. However, the leading varieties with respect to total yield of dry matter and protein per acre were from the medium maturity class. The medium maturity class displayed the best seasonal distribution of dry matter and general adaptation.

INTRODUCTION

Orchardgrass (*Dactylis glomerata* L.) is the most important forage grass in the Lower Mainland of British Columbia. The variety Danish is most commonly used and to a lesser degree S 143. These are medium-early and late-maturing varieties respectively.

The demand in many regions is for improved late varieties because they do not compete severely with associated legumes (10) are high in protein (2, 9), make good recovery growth (4, 5, 10), and produce a greater number of tillers than early varieties (4, 5, 6). According to Kelly's postulate (6), the greater number of tillers should enable them to produce greater mid-season dry matter yields than the early varieties. The major shortcomings of the late varieties are that they produce relatively low total yields of dry matter and of protein per acre (5, 9).

The purpose of the present study was to investigate the relations between maturity class and various other characteristics of orchardgrass under a silage-pasture type of utilization.

MATERIALS AND METHODS

Forty orchardgrass varieties representing a wide range in maturity and source (Table 1) were seeded on May 25, 1956, on Monroe silt loam soil at the Experimental Farm, Agassiz, B.C. The experimental design was a randomized block with five replications. Each plot consisted of 5 rows, 7 inches apart and 20 feet long. The seeding rate was 18 pounds of viable seed per acre.

During the preparation of the seed bed, 10 tons per acre of manure, 2 tons per acre of ground limestone, and 400 pounds per acre of 10-20-10 fertilizer were applied to the plot area. Supplemental chemical fertilizer was applied yearly: 200 pounds per acre of 10-20-10 fertilizer in early March, and 100 pounds per acre of 33.5-0-0 fertilizer following the second and third clippings.

TABLE 1. — SEED SOURCE AND MATURITY CLASSIFICATION OF ORCHARDGRASS VARIETIES: NUMBER OF DAYS FROM MARCH 1 TO ONE-THIRD OF THE EARS FULLY EMERGED FROM THE SHEATH

(Average number of days for five replications and 3 years)

Variety	Seed source	Maturity group	No. of [1] days
L-926	Canada	Very early	63.3
L-925	Canada	Very early	63.4
L-924	Canada	Very early	63.5
L-932	Canada	Very early	63.6
Avon	Canada	Very early	64.7
Potomac	U.S.A.	Early	66.1
Palestine	U.S.A.	Early	66.2
Trogdon	U.S.A.	Early	67.1
M2-11142-53	U.S.A.	Early	67.3
Pa. Early Syn.	U.S.A.	Early	67.3
Wisc. 52	U.S.A.	Early	67.3
Akaroa	U.S.A.	Early	68.0
Oron	Canada	Early	68.1
I.O.G. 6	U.S.A.	Early	68.5
Akaroa	N.Z.	Early	68.5
I.O.G. 1	U.S.A.	Medium	68.8
Pajbjerg II	Denmark	Medium	68.9
Commercial No. 1 [2]	Denmark	Medium	68.9
Commercial No. 1	Denmark	Medium	69.0
Skandia II	Sweden	Medium	69.2
Pa. Medium Syn.	U.S.A.	Medium	69.7
Coxa	Sweden	Medium	70.1
Roskilde II	Sweden	Medium	70.3
Milka II	Denmark	Medium	70.6
Gullaker	Sweden	Medium	70.7
Hercules	Canada	Medium	71.1
Roskilde Late II	Sweden	Medium	71.1
Tardus II	Sweden	Medium	71.1
Barenza	Holland	Medium	71.2
Hercules	Canada	Medium	71.2
Frode	Sweden	Medium	71.3
Utah Syn. 2	U.S.A.	Medium	71.8
S-37	Britain	Medium	71.9
S-143	Britain	Late	73.5
Pa. Late Syn.	U.S.A.	Late	75.1
P-2453 (Latar)	U.S.A.	Late	76.0
S-26	Britain	Very late	78.8
Ott. 100	Canada	Very late	79.3
Syn. F (Cornell)	U.S.A.	Very late	79.5
Agassiz PX 55	Canada	Very late	79.8
Mean			70.0
S.E. Mean			0.7

[1]Any two means not enclosed by the same bracket are significantly different at the 5 per cent level (3)
[2]Hereafter designated as "Danish"

During the three harvest years the characteristics listed below were studied.

Maturity Class: The number of days, from March 1, required by each variety to subtend one-third of its inflorescence free of the sheath.

Yield of Dry Matter: The first harvest each year was made at the stage when maturity was assessed, and subsequent clippings when the forage reached a height of 12 to 14 inches. Plots were cut on an individual basis

TABLE 2. — THE AVERAGE YIELD OF DRY MATTER[1] AND PROTEIN[2] IN POUNDS PER ACRE FOR 40 ORCHARDGRASS VARIETIES[3]

Variety	Dry matter	Variety	Protein
Gullaker	5491	I.O.G. 1	859
I.O.G. 1	5230	L-925	843
Avon	5153	Avon	840
Potomac	5137	Gullaker	825
I.O.G. 6	5090	L-932	823
Trogdon	5087	Potomac	796
Pa. Medium Syn.	5086	Skandia II	793
L-925	5080	Akaroa	793
M2-11142-53	5077	L-924	785
Oron	5033	Danish	784
Danish	4957	L-926	779
Hercules	4937	Trogdon	778
Pajbjerg II	4926	Pa. Early Syn.	776
Danish	4879	Hercules	773
Skandia II	4875	M2-11142-53	758
Pa. Early Syn.	4854	I.O.G. 6	756
Akaroa	4836	Oron	752
Coxa	4827	Wisc. 52	746
Wisc. 52	4822	Danish	745
Frode	4817	Roskilde Late II	739
L-932	4809	Hercules	738
Roskilde II	4793	Pajbjerg II	738
Hercules	4773	Akaroa	736
Roskilde Late II	4705	Barenza	734
Utah Syn. 2	4698	Milka II	733
S-26	4683	Coxa	732
Barenza	4677	Frode	731
Pa. Late Syn.	4671	Pa. Medium Syn.	728
Akaroa	4653	Agassiz PX. 55	715
Tardus II	4650	Tardus II	709
L-924	4623	S-26	709
Milka II	4611	Pa. Late Syn.	701
S-37	4478	Roskilde II	699
L-926	4476	Ott-100	696
Syn. F (Cornell)	4227	Utah Syn. 2	688
S-143	4221	S-143	681
P-2453 (Latar)	4089	S-37	678
Agassiz PX. 55	4089	Syn. F (Cornell)	651
Ott-100	4027	P-2453 (Latar)	644
Palestine	3555	Palestine	534
Mean	4743		742
S.E. Mean	166.6		13.1

[1]Average yield of dry matter for 3 years
[2]Average yield of protein for 1957 and 1959
[3]Any two means not enclosed by the same bracket are significantly different at the 5 per cent level (3)

when they reached the prescribed height using a sickle mower adjusted to leave a 4-inch stubble. Immediately following clipping a 500-gram sample was taken from each plot for dry matter determinations. These samples were dried at 100°C., ground to a fineness of a 100 mesh to the inch screen, and analysed for protein N x 6.25 (1). Samples waiting drying were held at −17°C.

Density: This was determined yearly on all plots following the second clippings. It was based on the number of tillers for one metre of row taken at random within the plot area.

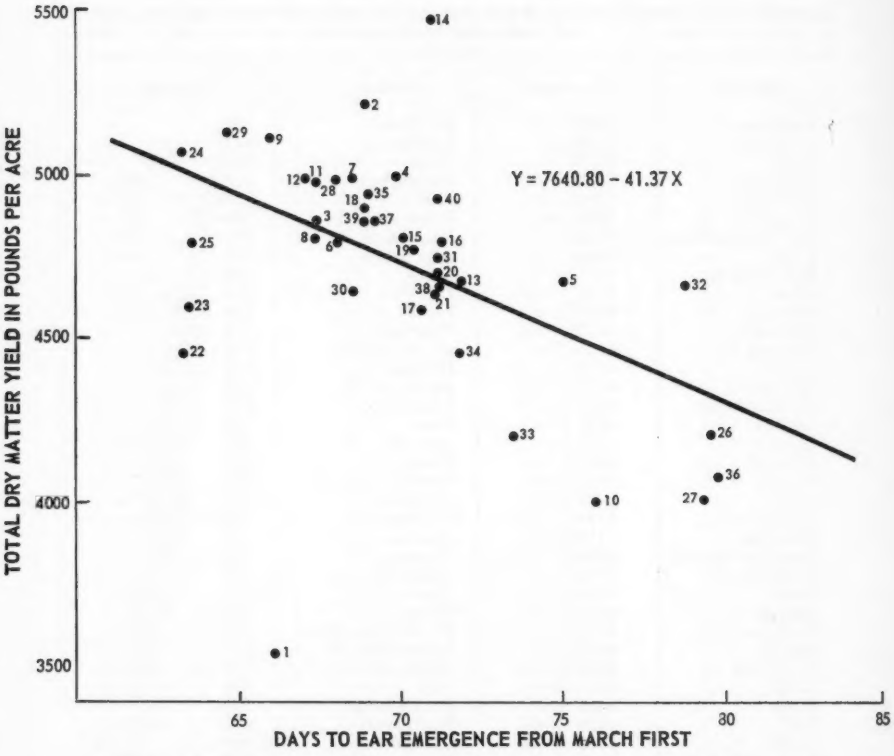

FIGURE 1. The effect of maturity upon the total yield of dry matter for 40 orchardgrass varieties, mean data for five replications and 3 years, 1957-1959.

Dormancy: Fall dormancy and its opposite characteristic fall greenness were rated on the basis of 1-9, with 1 indicating complete dormancy and 9 fully green.

RESULTS

In the establishment year good stands were obtained for all varieties. Throughout the 4 test years winter injury was nil, with the exception of Palestine which received slight injury.

Maturity

There was no significant interaction between years and the number of days to heading. The data in Table 1 show 70 days as the mean number of days for one-third emergence. The data also show a difference of 16.5 days between the earliest and the latest variety for this factor. On the basis of the data in Table 2, five arbitrary maturity groups or classes were established: *very early*, *early*, *medium*, *late*, and *very late*. Differences among group means were significant except between adjacent groups.

TABLE 3. — THE RELATIVE ORDER OF MATURITY GROUPS WITH RESPECT TO THE INTERVAL BETWEEN CUTS

Cutting period	Maturity groups in ascending order of number of days between cuts
May-June:	late < very late < early < medium < very early
June-July:	early = medium < very late < late < very early
July-Aug:	medium = late = very late < very early < early
July-Sept:	early = late < medium < very late < very early
Sept.-Oct.:	very early < very late < medium < early = late

TABLE 4. — THE RELATIVE ORDER OF MATURITY GROUPS WITH RESPECT TO YIELD FOR INDIVIDUAL CUTS

Cutting period	Maturity groups in descending order of yield of dry matter
May-June:	very early > very late > early > medium > late
June-July:	medium > very early > early = very late > late
July-Aug:	medium > early = very late > very early = late
Aug.-Sept:	medium = early > late > very late > very early
Sept.-Oct:	medium > early > late > very late = very early

TABLE 5. — THE RELATIVE ORDER OF MATURITY GROUPS WITH RESPECT TO YIELD OF DRY MATTER FOR A CALENDAR PERIOD

Growth period	Maturity groups in descending order of yield of dry matter
April	very early > early > medium > late > very late
May	late > very late > early > medium > very early
June	medium > early > very late > late > very early
July	medium > very late > late > early = very early
August	medium = late > very early > early > very late
September	very early > medium > early > very late > late

The yield data for dry matter and protein per acre are given in Table 2. The highest yield of dry matter for the 3 harvest years was produced by the variety Gullaker. Considering the mean yield of dry matter, 80 per cent of the early group, 67 per cent of the medium group, and 60 per cent of the very early group were above the mean. All varieties in the late and very late group were below the mean. The regression of dry matter on maturity in Figure 1, where a highly significant correlation of $r = -.47$ ($n = 40$) was obtained, indicates that high total yields are associated with early maturity. The lack of conformity of Palestine to the general distribution indicates its low adaptation to this region. With Gullaker the opposite is true and its lack of conformity to the general distribution indicates its very high yield potential and adaptation to this region.

The data in Figure 2 give the seasonal distribution of dry matter based on the average of the 1957 and 1959 seasons. Data for the 1958 season were omitted because no cuttings were made of any of the varieties after the first week in July, due to severe drought. Each of the five maturity groups required a different number of days to reach the prescribed cutting height of 12 to 14 inches. Their relative order in this respect is given in Table 3.

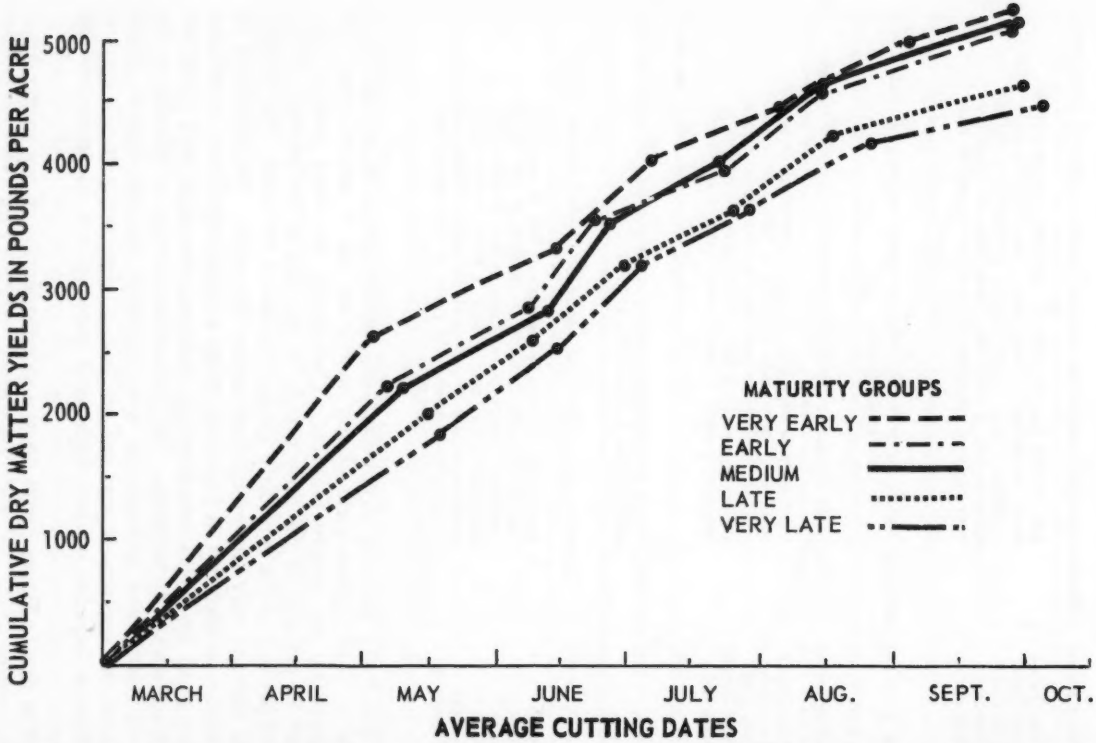

FIGURE 2. The average cumulated dry matter yields of five maturity groups of orchardgrass cut six times in 1957 and 1959. Mean data for five replications.

The relative order of the five maturity groups with respect to yield of dry matter is given in Table 4. The change in the order of the groups from that given in Table 3 indicates that yield does not parallel rapidity of recovery where cutting is conducted on a height basis.

The cutting intervals indicated in Figure 2 are based on a growth stage and are not confined to a calendar period or to the same period for the five maturity groups. To compare yield or growth the assessment should be over the same period of a season. In Table 5 the relation between maturity groups for yield of dry matter is given for a calendar period, based on the data in Figure 2. These data indicate the early and late season potential of the very early group, the favourable mid-season potential of the medium group, and the high potential of the late and very late group for the May period, the only season in which these two groups predominate.

The data for protein in Figure 3 show wide fluctuations in the levels of this constituent over the six cutting periods. The low level of all groups for Cut 1 is due to the much later growth stage (heading) at which this cut was made. Nitrogen was applied following Cuts 1 and 3. However, because cutting was done on an individual basis, it was impossible to apply fertilizer on a calendar date. Rain falling before or after a particular application may have accentuated the fluctuations encountered in the protein levels. The data show a marked similarity between the very early and very late maturity groups for both protein content and seasonal response, a slightly lower degree of similarity between the early and late group, and the lower protein content, with the exception of the Cut 3 period, for the medium maturity group.

The yield data in Table 2 show wide varietal differences in yield of protein per acre. The considerable difference of 325 pounds of protein per acre was obtained between the high and low variety. A highly significant negative correlation, $r = -.52$ ($n = 40$) was obtained between yield of protein and maturity. In general, total yield of dry matter was the main determinant for protein yield.

The yearly assessments of tillering and fall dormancy were highly uniform and there was no significant variety x year interaction. The tillering data in Table 6 for 1959, the final test year, show significant differences between certain varieties. However, these differences were independent of maturity group. In the investigation of association between characteristics (Table 7) no relation was found between tillering and per cent protein, maturity or yield. A significant negative correlation was obtained between tillering and fall dormancy.

The data in Table 6 indicate significant relations between dry matter yields and days to heading, per cent protein and fall dormancy, tillering and fall dormancy. Prostrate fall growth habit, not previously reported in the data, was positively associated with fall dormancy. A significant correlation of $r = +.69$ ($n = 40$) was obtained between prostrate or rosette type of fall growth and fall dormancy.

TABLE 6. — TILLERING AND FALL DORMANCY OF 40 ORCHARDGRASS VARIETIES MEAN VALUES[1] FOR FIVE REPLICATIONS

Variety	Tillers[2] per 1-metre row	Variety	Fall dormancy (1-9)[3]
S-37	318	L-924	1.6
Avon	293	P 2453 (Latar)	1.8
I.O.G. 6	287	L-932	1.8
S-143	283	L-926	2.0
Utah Syn. 2	281	L-925	2.0
Ottawa 100	277	Syn. F (Cornell)	2.0
Barenza	277	Avon	2.2
Trogdon	269	I.O.G. 1	3.0
Roskilde Late II	263	M2-11142-53	3.0
Danish	263	I.O.G. 6	3.2
Milka II	260	Ottawa 100	3.2
Akaroa	257	Pa. Late Syn.	3.4
Roskilde II	255	Wisc. 52	3.4
S-26	255	Trogdon	3.4
Hercules	255	Akaroa	3.6
Akaroa	252	Agassiz PX 55	3.6
Skandia II	251	Hercules	4.0
Pa. Medium Syn.	246	Tardus II	4.2
Potomac	246	Oron	4.4
Frode	244	Pa. Early Syn.	4.6
L-924	243	Roskilde II	4.8
Hercules	242	Potomac	5.0
L-925	241	Gullaker	5.0
L-926	239	Frode	5.0
Coxa	237	Danish	5.0
Danish	234	Utah Syn. 2	5.4
I.O.G. 1	233	Pajbjerg II	5.4
Gullaker	232	Roskilde Late II	5.4
Palestine	231	Skandia II	5.4
Agassiz PX 55	231	Danish	5.4
Oron	229	Coxa	6.0
L-932	227	Milka II	6.0
Wisc. 52	225	Pa. Medium Syn.	6.2
Tardus II	218	Hercules	6.4
P 2453 (Latar)	216	Palestine	6.6
Pa. late Syn.	212	Akaroa	7.0
M2-11142-53	211	S-37	7.0
Pa. Early Syn.	203	Barenza	7.2
Syn. F (Cornell)	203	S-26	7.8
Pajbjerg II	202	S-143	8.0
Mean	246		4.5
S.E. Mean	16.7		.36

[1]Any two means not enclosed by the same bracket are significantly different at the 5 per cent level (3)
[2]1959 assessment. Counts taken following 2nd clipping
[3]November 1959 assessment. 1 dormant, 9 winter green

DISCUSSION

The 40 orchardgrass varieties assessed in this investigation covered a wide range of plant material. The average difference of 16.5 days between the earliest and latest varieties in the number of days required for ear emergence indicates a major advantage for the early varieties in respect of availability of aftermath for grazing following a silage harvest at the recommended stage of ear emergence. The partitioning of the varieties into arbitrary maturity groups or classes is supported to a high degree by the

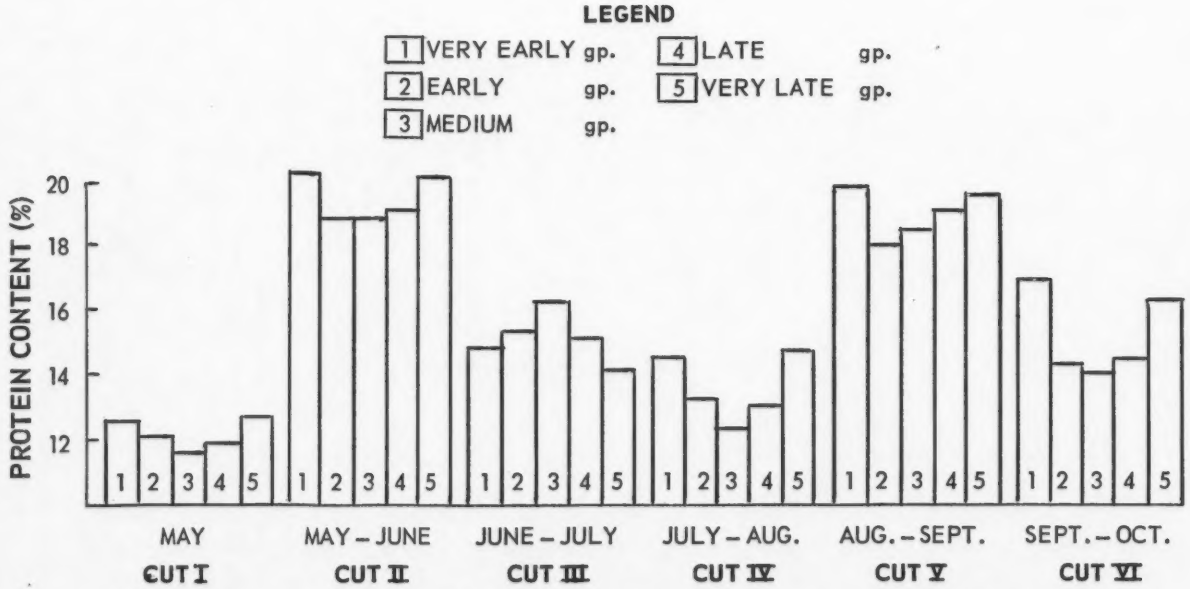

FIGURE 3. Seasonal protein levels for five maturity groups of orchardgrass harvested six times in 1959.

TABLE 7. — SIMPLE CORRELATION COEFFICIENTS FOR FIVE GROWTH CHARACTERISTICS OF ORCHARDGRASS VARIETIES

	Days to heading	Per cent protein	Tillering	Fall dormancy
Yield of dry matter	−.47**	.00	+.04	−.06
Days to heading		−.02	+.01	+.22
Per cent protein			−.08	+.31*
Tillering				−.40**

*P exceeds 0.05, i.e. .30 where n = 40
**P exceeds 0.01, i.e. .39 where n = 40

data presented. The small number of varieties in the extreme groups reduced the reliability of the investigations with respect to those groups.

The observations of tillering indicated that number of tillers was not associated with maturity groups or yield of dry matter. The absence of association between tillering and yield does not agree with the findings of Kelly (6). This does not exclude the possibility that tillers are associated with yield of dry matter on a different basis than in respect of numbers and it is possible, as suggested by Langer (7), that the association may be between thickness of tillers. This points to the need for further invesigation.

Fall dormancy was associated with high per cent protein, low number of tillers, and prostrate fall growth. An indefinite relation was indicated with maturity and no relation with yield. As to which is preferable, fall dormancy or winter greenness, any decision would depend upon the region and time of utilization. Early dormancy could be desirable in a region with severe fall and winter conditions but undesirable under temperate conditions where late fall or winter grazing was the practice.

Considering dry matter and protein, the very early group was outstanding in the early spring but did not display a satisfactory seasonal distribution in later cuts though per cent protein was high throughout. The early group was intermediate for seasonal distribution but produced good total yields of dry matter with only two varieties yielding less than the mean. The very late group was very slow in spring growth and low in total yield of dry matter but high in per cent protein. The best production of this group was in May and July but in no month was it the leading group. The late group reacted similarly to the very late group. It was the highest for yield of dry matter in May and ranked with the medium group in August. In September the late groups were the lowest for production of dry matter. All varieties from both the late and very late groups were below the mean for total yield of dry matter.

The outstanding varieties for total yield of dry matter, yield of protein per acre and seasonal distribution were from the medium maturity group, indicating the adaptation of this maturity class for the Lower Mainland region of British Columbia. However, individual varieties with superior characteristics were found within each maturity group, thus indicating the potential usefulness of these varieties in breeding programs.

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THE PERSISTENCE OF CERTAIN SOIL INSECTICIDES FOR CONTROL OF THE TUBER FLEA BEETLE, *EPITRIX TUBERIS* (GENT.), IN THE INTERIOR OF BRITISH COLUMBIA[1]

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ABSTRACT

In 1953, dieldrin, aldrin and chlordane were applied to potato plots to control the tuber flea beetle. The insecticides were applied at 1.5, 4 and 10 pounds of toxicant per acre, respectively, and harrowed or rotary-tilled into the soil. Aldrin harrowed and chlordane rotary-tilled into the soil provided satisfactory control (i.e., 80 per cent or more of marketable tubers) in 1954 and 1955, whereas dieldrin rotary-tilled and chlordane harrowed into the soil provided less effective but acceptable control (i.e., 70 to 80 per cent of marketable tubers) for the same years. In 1956, all treatments significantly reduced damage but not so from a practical standpoint. In 1957, dieldrin, aldrin and chlordane were applied at one-quarter, one-half and the full 1953 rates to half of the plots treated in 1953 and to previously untreated plots. For all insecticides, increasing the rate of application significantly increased the degree of control; the one-half and full rates continued the above levels of control to 1958. Active residues from each of the insecticides remained in the soil from 1953 to 1958.

INTRODUCTION

Various workers have shown the effectiveness of soil treatments of chlorinated hydrocarbon insecticides such as aldrin[2], chlordane[3] and dieldrin[4] applied against the tuber flea beetle (1, 4, 7). In the southern interior of British Columbia, this work has led to the widespread and continued use of these treatments. Many workers (2, 3, 5, 6) have reported that these and other chlorinated hydrocarbon insecticides persist for long periods when applied to the soil. This is a report on studies conducted at Kamloops from 1953 to 1958 on the relative effectiveness and persistence of these insecticides when applied against the tuber flea beetle in the southern interior of British Columbia, and also on the effects of two commonly used cultural practices on these factors.

MATERIALS AND METHODS

In May 1953, aldrin, chlordane and dieldrin were applied as soil treatments to plots of uncontaminated soil at rates previously found to be effective against the tuber flea beetle (1). Plots of 1/70-acre were replicated four times in a randomized block design. The treatments were as follows:

1. Aldrin, 2½ per cent dust, applied at 4.0 pounds of toxicant per acre and rotary-tilled to a depth of 6 inches.
2. The same insecticide at the same rate of application harrowed to a depth of 2 to 3 inches.
3. Dieldrin, 1½ per cent dust, at 1.5 pounds and rotary-tilled to a depth of 6 inches.
4. Chlordane, 5 per cent dust, at 10.0 pounds and rotary-tilled to a depth of 6 inches.
5. The same insecticide at the same rate of application harrowed to a depth of 2 to 3 inches.
6. Untreated.

[1]Contribution No. 13, Entomology Laboratory, Research Branch, Canada Department of Agriculture, Kamloops, B.C.
[2]1, 2, 3, 4, 10, 10-hexachloro-1, 4, 4a, 5, 8, 8a-hexahydro-1, 4-endo-exo-5, 8-dimethanonaphthalene.
[3]1, 2, 4, 5, 6, 7, 8, 8-octachloro-4, 7-methano-3a, 4, 7, 7a-tetrahydroindane.
[4]1, 2, 3, 4, 10, 10-hexachloro-6, 7-epoxy-1, 4, 4a, 5, 6, 7, 8, 8a-octahydro-1, 4-endo-exo-5, 8-dimethanonaphthalene.

The insecticides were applied with hand shakers made from 1-pound tobacco tins with perforated tops. These were agitated within 12 inches of the soil surface to minimize drifting of the insecticide. Immediately after application the insecticides were incorporated into the plots with either a tractor-mounted, 6-foot, Howard Rotovator or a tractor-drawn, diamond-tooth harrow. To ensure thorough incorporation, the plots were cultivated in two directions at right-angles to one another.

After treatment the experimental plots were summer-fallowed and frequently harrowed to eradicate a heavy infestation of couch grass (*Agropyron repens* (L.) Beauv.). In 1954, 1955, and 1956, the plots were machine-planted to Foundation "A", Netted Gem potatoes. After 1953, no deep tillage was practised, other than that resulting from machine-planting and harvesting each spring and fall. In all other respects the plots received normal cultural treatment, including sprinkler irrigation.

In 1954, 1955, and 1956, effectiveness of the treatments was appraised at harvest by counting larval tunnels in the tubers from 20-pound sub-samples of marketable-sized tubers. The sub-samples were selected at random from 24-hill samples taken from each plot. These hills were taken on a diagonal line ending 3 feet or more from the plot boundaries. Each tuber in the sub-sample was hand-peeled and the number of larval tunnels recorded according to the following groupings: 0, 1-4, 5-9, 10-14, 15-19, 20 and over. Tubers having 9 or fewer larval tunnels were considered to be marketable, while those having 10 or more were not. In terms of economic control, tuber flea beetle control treatments are considered to be satisfactory if they provide 80 to 100 per cent marketable tubers when assessed by the previously described method. In farm practice this standard is overly rigid. Treatments which provide 70 to 80 per cent of marketable tubers are considered to provide economic control. Although 70 per cent may appear low, the criterion for determining marketable tubers in these experiments is very strict (viz., 9 or fewer larval tunnels per tuber)*. Counts of over 300 larval tunnels are not uncommon in heavily damaged tubers.

In May, 1957, two of the four randomized blocks were abandoned and two additional blocks were set out in uncontaminated land immediately adjacent to those retained. Except for application rate and formulation, each of the retained plots received similar insecticide and cultural treatments in 1953 and in 1957. The added blocks also received comparable insecticide and cultural treatments. Each plot was divided into three 1/210-acre sub-plots. One of these received one-quarter of the rate of the particular insecticide that was applied in 1953, one received one-half, and the third the full rate; the three sub-plot treatments were randomized. Emulsifiable concentrates of aldrin 20 per cent, chlordane 65.5 per cent, or dieldrin 20 per cent, were used in place of the dust formulations. The insecticide for each sub-plot was diluted with water to make 2 gallons and applied with a watering-can. As in 1953, immediately after application the insecticides were incorporated into the soil by rotary-tilling or harrowing.

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*Marketable sized tubers may not have a minimum diameter of less than 1½ inches.

The 1957 treatments were appraised in 1958 only. A very light infestation of tuber flea beetles in 1957 produced inconclusive results. In all other respects this experiment was similar to that conducted from 1953 to 1956.

The results were statistically analysed by first transforming the percentages to degrees by the formula : angle = arc sin $\sqrt{\text{percentage}/100}$. In the summary tables, the standard errors refer to the transformed values only. The mean percentages presented in the tables have been derived from the means of the transformed values.

RESULTS AND DISCUSSION

In 1954, 1955 and 1956, the percentage of marketable tubers was greater in the plots which received insecticide in 1953 than in the untreated plots. The differences reached at least the 5 per cent level of significance each year, although over the same period the general level of flea beetle damage in the plots increased.

Results for the various insecticide treatments followed much the same pattern in the 3 years, but by 1956 the differences between them were no longer significant ($P > 0.05$). Aldrin was more effective when harrowed into the soil than when rotary-tilled; conversely, chlordane was more effective when rotary-tilled than when harrowed, but the differences for this insecticide were less conclusive.

In terms of economic control the harrowed aldrin and rotary-tilled chlordane treatments provided satisfactory control (i.e., 80 per cent or more of marketable tubers) in 1954 and 1955. The harrowed chlordane and rotary-tilled dieldrin treatments were acceptable (i.e., 70 to 80 per cent of marketable tubers) in the same years. None of the treatments was economically acceptable in 1956.

Most of the insecticide treatments which were applied in 1957 reduced tuber flea beetle damage in 1958. Except for the previously untreated plots

TABLE 1.—MEAN PERCENTAGES[1] OF MARKETABLE TUBERS[2] IN 1954, 1955 AND 1956 FOLLOWING APPLICATION IN 1953 OF ALDRIN, DIELDRIN AND CHLORDANE BY TWO METHODS

Treatment	Toxicant per acre, lb.	Mean percentages from transformed values[3]			Means of percentages transformed to degrees[1]		
		1954	1955	1956	1954	1955	1956
Aldrin, 2½% dust: Harrowed	4	88.0	87.7	25.9	69.7	69.5	30.6
Rotary-tilled	4	58.9	69.1	12.0	50.1	56.2	20.3
Chlordane, 5% dust: Harrowed	10	74.1	81.1	26.2	59.4	64.2	30.8
Rotary-tilled	10	84.0	95.5	16.7	66.4	77.7	24.1
Dieldrin, 1½% dust: Rotary-tilled	1.5	79.7	77.1	12.6	63.2	61.4	20.8
Untreated	—	34.4	19.5	0.5	35.9	26.2	4.1
S.E.					±4.2	±3.6	±3.9

[1]Transformed by the formula: angle = arc sin $\sqrt{\text{percentage}/100}$
[2]Of marketable size and having 9 or fewer larval tunnels
[3]Derived back from means of transformed percentages

which were treated with chlordane at either of the two lower rates in 1957, the differences from the untreated plots were highly significant ($P < 0.01$).

Higher percentages of marketable tubers ($P < 0.01$) resulted for each insecticide when the application rates were increased from 1 to 2 to 4 pounds of toxicant per acre for aldrin, from 2.5 to 5 to 10 pounds for chlordane or from 0.38 to 0.75 to 1.5 pounds for dieldrin. This was most evident in the plots which were treated with chlordane in 1957 only. The over-all reduction of tuber flea beetle damage was more marked in the plots which were treated with insecticides in both 1953 and 1957, particularly for the chlordane-treated plots ($P < 0.05$).

Aldrin appeared to be more effective when incorporated into the soil by harrowing rather than by rotary-tilling; this was true of plots treated in 1953 and 1957 and in 1957 only ($P < 0.05$). Chlordane applied to plots in both 1953 and 1957 tended to be more effective when rotary-tilled than when harrowed, but this was not so for the plots treated with chlordane only in 1957.

In 1958, in terms of economic control, aldrin applied to plots only in 1957, at 4 pounds of toxicant per acre, gave satisfactory control when

TABLE 2.—PERCENTAGES OF MARKETABLE TUBERS[1] IN 1958 FOLLOWING INSECTICIDE APPLICATIONS IN 1953 AND 1957, OR IN 1957 ONLY, GIVING MEANS FOR ALDRIN, DIELDRIN AND CHLORDANE, RATES AND METHODS OF APPLICATION

Treatment	Toxicant per acre, lb.		Mean percentages from transformed values[2]	
	1953	1957	Treated in 1953 and 1957	Treated in 1957 only
Aldrin harrowed	4	1	65.8	57.6
	4	2	87.8	58.2
	4	4	92.4	100.0
Aldrin rotary-tilled	4	1	58.0	44.8
	4	2	60.4	42.4
	4	4	81.1	75.3
Chlordane harrowed	10	2.5	35.8	1.4
	10	5	79.4	9.5
	10	10	84.4	66.8
Chlordane rotary-tilled	10	2.5	53.2	7.6
	10	5	87.4	12.4
	10	10	99.3	63.2
Dieldrin rotary-tilled	1.5	0.38	42.5	33.6
	1.5	0.75	73.2	42.0
	1.5	1.5	86.2	78.6
Untreated	—	—	1.7	6.5

[1]Of marketable size and having 9 or fewer larval tunnels

[2]Derived back from means of transformed percentages (means of percentages transformed by the formula: angle = arc sin $\sqrt{\text{percentage}/100}$

harrowed into the soil. The same insecticide and rate, rotary-tilled, gave acceptable control as did dieldrin at 1.5 pounds, also rotary-tilled. The other treatments were economically unacceptable.

As previously noted, there were higher percentages of marketable tubers in the plots which were treated with insecticide in both 1953 and 1957. Satisfactory control resulted following applications in 1957 of aldrin at rates as low as 2 pounds of toxicant per acre harrowed into the soil and of chlordane at rates as low as 5 pounds rotary-tilled into the soil.

By 1956, the insecticides applied in 1953 had become economically unacceptable. The possible explanation that insect resistance had developed to any appreciable degree is remote because further applications to the experimental plots in 1957 restored their effectiveness against the tuber flea beetle. In addition, continual checks of treatment effectiveness in the field gave no indication of the development of partial or complete resistance to any of the chlorinated hydrocarbons nor has this been substantiated by reports from other areas where this insect is an economic pest.

Insecticidally-active residues from aldrin, chlordane and dieldrin remained in the soil at least until 1958, 5 years after application. Although predictions cannot be based entirely on the results of this experiment, it is probable that with climatic and soil conditions similar to those under which this work was conducted, one application of aldrin at 4 pounds of toxicant per acre harrowed into the soil should provide satisfactory control against the tuber flea beetle for three consecutive seasons. A supplementary application of not less than 2 pounds in the fourth season should continue this high level of control to at least the fifth season. Likewise, one application of dieldrin at the 1.5-pound rate rotary-tilled into the soil should provide acceptable control for three consecutive seasons. A supplementary application of not less than 0.75 pounds in the fourth season should continue this control to at least the fifth season. Although somewhat less reliable because of inexplicably poor control results in 1958 following application in 1957, chlordane at the 10-pound rate incorporated into the soil by rotary-tilling should also provide satisfactory control for three consecutive seasons. A supplementary application of not less than 5 pounds in the fourth season should continue this control for another season. Similarly, the same insecticide at the 10-pound rate harrowed into the soil should provide acceptable control for three consecutive seasons and, after a supplementary application of not less than 5 pounds in the fourth season, should continue this level of control for another season.

It is obvious that substantial savings can be made if two applications of insecticide will provide satisfactory control against the tuber flea beetle for at least five seasons. The total cost of insecticides for two applications at the suggested rates is $22.00 to $27.00 per acre for five seasons instead of $15.00 to $18.00 per acre each season at the previously recommended rate. A substantial saving in the use of labour and machinery would also result. The persistence of aldrin, chlordane and dieldrin in the soil for periods longer than one growing season provides a further advantage. Soil-inhabiting insects such as wireworms, white grubs, and certain cutworms are controlled in years of a crop rotation when potatoes are grown as well as

TABLE 3.—PERCENTAGES OF MARKETABLE TUBERS[1] IN 1958 FOLLOWING INSECTICIDE APPLICATIONS IN 1953 AND 1957, OR IN 1957 ONLY, GIVING MEANS OF ONE AND TWO APPLICATIONS MADE BY TWO METHODS

	Method of incorporation	Mean percentages from transformed values[2]				Means of percentages transformed to degrees[3]			
		Aldrin	Chlordane	Dieldrin	Untreated	Aldrin	Chlordane	Dieldrin	Untreated
Treated in 1953 and 1957	Harrowed	83.4	67.9	—	—	65.9	55.5	—	—
	Rotary-tilled S.E.	66.9	84.9	68.5	—	54.9	67.1 ±6.39	55.9	—
	Mean (and S.E.)	75.2	76.4	68.5	1.7	60.4 (± 4.52)	61.3 (± 4.52)	55.9 (± 6.39)	7.6 (± 6.39)
Treated in 1957 only	Harrowed	79.4	20.0	—	—	63.0	26.6	—	—
	Rotary-tilled S.E.	54.5	24.6	51.9	—	47.6	29.7 ±6.39	46.1	—
	Mean (and S.E.)	67.0	22.3	51.9	6.5	55.3 (± 4.52)	28.2 (± 4.52)	46.1 (± 6.39)	14.8 (± 6.39)

[1]Of marketable size and having 9 or fewer larval tunnels
[2]Derived back from means of transformed percentages
[3]Transformed by the formula: angle = arc sin $\sqrt{\text{percentage}/100}$

TABLE 4.—PERCENTAGES OF MARKETABLE TUBERS[1] IN 1958 FOLLOWING INSECTICIDE APPLICATIONS IN 1953 AND 1957, OR IN 1957 ONLY, GIVING MEANS FOR THREE RATES APPLIED IN 1957

	Mean percentages from transformed values[2]				Means of percentages transformed to degrees[3]			
	Plots treated 1953 and 1957	Plots treated 1957 only	Mean	Net effect 1953 treatment	Plots treated 1953 and 1957	Plots treated 1957 only	Mean	Net effect 1953 treatment[4]
¼ 1953 rate	51.0	25.0	38.0	30.8	45.6	29.9	37.8	22.9 ($P = 0.05$)
½ 1953 rate	78.4	31.1	54.8	52.1	62.3	33.9	48.1	35.6 ($P < 0.01$)
Full 1953 rate	90.1	80.8	85.5	14.1	71.7	64.0	67.8	14.9 ($P > 0.05$)
S.E. for vertical comparisons					±3.61	±3.61	±2.55	±5.10
Untreated	1.7	6.5	4.1		7.6	14.8	11.2	

[1]Of marketable size and having 9 or fewer larval tunnels
[2]Transformed mean percentages (viz. 3) "de-transformed"
[3]Transformed by the formula: angle = arc sin $\sqrt{\text{percentage}/100}$
[4]Differences between blocks have been eliminated in calculating the net effect of the 1953 treatment, by taking differences from the results for untreated plots.

when they are not (1). The use of low-rate supplementary treatments at intervals of 2, 3 or 4 years instead of annual full-rate treatments may help to prevent serious accumulations of toxic residues in land which is being used continually for potatoes or other crops requiring treatment of the soil with the same or other chlorinated hydrocarbon insecticides. It must be emphasized that, under field conditions, very different from those under which this experiment was conducted, modifications may have to be made in the rates of the supplementary treatments and intervals at which they must be applied.

Additional work is necessary to establish whether or not it is feasible to apply supplementary treatments of different chlorinated hydrocarbons rather than the original insecticide in order to maintain a minimal toxicity level in the soil. Also, a simple method is required for determining the amount and timing of supplementary applications to maintain this level. These factors will be difficult to determine and likely will be specific for each particular instance. Mainly, they will be dependent upon the soil type and condition, cultural practices followed, and climatic conditions.

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NOTE ON BISMALEAMIC ACID AS A TOBACCO SUCKER GROWTH INHIBITOR[1]

The search for suitable means of inhibiting sucker growth on tobacco plants is a problem of primary concern. The most successful inhibitor to date, maleic hydrazide (MH), has been widely tested and is highly favoured among growers. However, doubts have arisen concerning the use of MH because of reported changes in constituent ratios (1, 2) which might impair the quality and increase resistance to its use by the tobacco manufacturers and importers. This conflict of grower's and manufacturer's interests has given new impetus to the search for growth regulators without these undesirable secondary effects.

A straight-chain compound bismaleamic acid (BMA) (HOOC-CH=CH-CO-NH-)$_2$ has been patented as a growth inhibitor (3). This chemical was applied on tobacco plants grown in greenhouse and was found to be an effective inhibitor of tobacco sucker growth. The yield of leaves was increased by use of this compound.

Tobacco seeds, variety Hicks, were germinated in sand and the seedlings were transplanted to soil in 6-inch pots. When the plants reached the flower bud stage, the inflorescence and the top four leaves were cut off and the plants were sprayed with aqueous solutions of both BMA and MH containing a few drops of Tween 20 to facilitate wetting of the leaves. The treatments were: 1) BMA, 1×10^{-4} moles; 2) BMA, 2×10^{-4} moles; 3) MH, 1×10^{-4} moles per plant; 4) an untreated check. The treatments used were calculated to approximate the amounts usually recommended in field use (2.25 pounds of MH per acre). Treatment 2 was included because preliminary growth experiments with soybeans indicated that aqueous solutions of BMA had a somewhat less strong inhibitory action than MH. There were five single plant replicates and they were arranged in a randomized block.

After 3 weeks, when the suckers on the check plants were well developed, leaves and suckers of all test plants were harvested and oven-dried. The average leaf and sucker weights are shown in Table 1.

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[1]Contribution No. 135 from Plant Research Institute, Canada Department of Agriculture, Ottawa, Ont. Contribution No. 15 from Analytical Chemistry Research Service, Canada Department of Agriculture, Ottawa, Ont.

TABLE 1. — AVERAGE WEIGHT (IN GRAMS) OF LEAVES AND SUCKERS FROM TOBACCO PLANTS TREATED WITH BISMALEAMIC ACID AND MALEIC HYDRAZIDE

	BMA 1×10^{-4}M	BMA 2×10^{-4}M	MH 1×10^{-4}M	Check	F	Necessary F	
						5%	1%
Leaves	13.60	12.27	12.89	9.74	3.53	3.49	5.95
Suckers	0.65	0.12	0.11	3.61	6.10	3.49	5.95

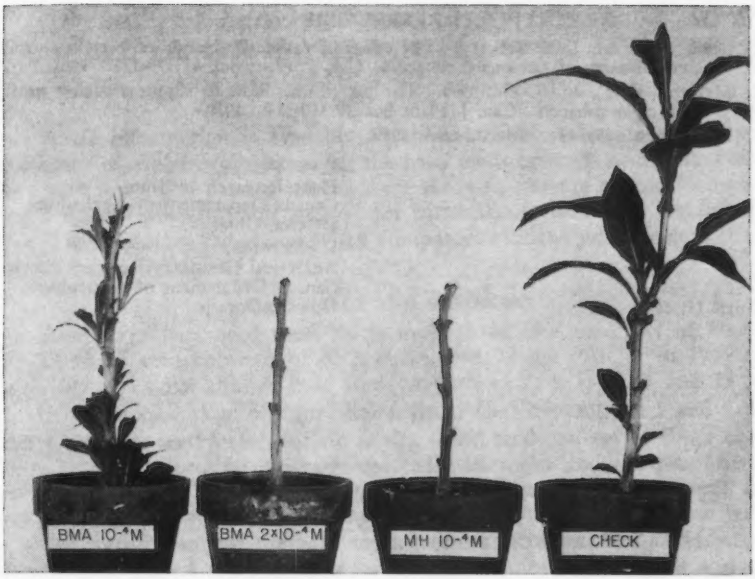

FIGURE 1. Sucker growth of four typical tobacco plants treated with BMA and MH after harvesting of leaves.

Statistical analysis of the data showed that for the weight of suckers there was: (a) a highly significant difference between the check and all other treatments; (b) a significant difference between treatment 1 and treatments 2 and 3. The differences in weight of leaves were not significant among treatments 1, 2 and 3, but there was significant difference between these three treatments and the check. The highest yield of leaves was obtained by treatment 1 and, although there was some growth of suckers, there appeared to be little advantage in using larger amounts of BMA.

Figure 1 shows four typical tobacco plants, which had been treated as outlined above, after harvesting of leaves. Marked differences in sucker growth are evident, with the check sample showing the greatest growth. The both BMA and the MH treatments show good sucker growth inhibition.

It is concluded that BMA with its sucker inhibiting action and increased leaf weight effect shows promise as a tobacco sucker growth inhibitor.

Work is continuing on the various ramifications of BMA treatments on tobacco.

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April 11, 1961

NOTE ON THE GROWTH CHARACTERISTICS OF ALFALFA AND RED CLOVER PLANTS DERIVED FROM HARD SEEDS AND SEEDS OF DIFFERENT SIZES[1]

Little information is available regarding the contribution to the performance of a plant population by the hard seeds or seeds of different sizes in a seed lot. The present studies were made to observe the performance, based on fall growth habit and winter survival, of these seed fractions in alfalfa and medium red clover when compared with the performance of the original seed lot.

MATERIALS AND METHODS

Studies of hard seed were made in 1958 on five seed lots of Vernal alfalfa (hard seed contents of 3, 3, 4, 14, and 33 per cent). In 1959, six seed lots of Vernal alfalfa (hard seed contents of 2, 3, 3, 4, 14, and 33 per cent), two seed lots of Ranger alfalfa (hard seed contents of 5 and 7 per cent), and one seed lot of Buffalo alfalfa (hard seed content of 7 per cent) were studied. Seed was soaked repeatedly in water and the germinating seed killed by drying until those that remained were ones that would not germinate in a 6-day test on blotting paper in petri dishes. Samples of each original seed lot and of the hard seeds from it were scarified and sown in the field in paired rows randomized in five replications in 1958 and ten replications in 1959.

Hard seeds from four seed lots of Wisconsin Common medium red clover (hard seed contents of 6, 8, 10, and 13 per cent) were obtained in the same manner as described above. Samples of each original seed lot and of the hard seeds from it were scarified and sown in 1959 in the field in paired rows randomized in four replications.

Seeds of different sizes were obtained from a seed lot of Vernal alfalfa produced in 1957 at Spooner, Wisconsin. During cleaning, the very smallest seed from the bottom screen during the first cleaning and the very largest seed from the top screen during the third cleaning were saved to compare with the original and final seed. The average weight of 100 seeds of the smallest seed was 0.1358 grams, of the largest seed 0.2757, of the original seed 0.2285, and of the final seed 0.2281 grams. Samples of each of the four seed fractions were sown in rows randomized in five replications in 1958 and in ten replications in 1959.

The alfalfa was seeded both years (1958 and 1959) in mid-May. The medium red clover was seeded on June 25, 1959, the only year it was sown. The clover was seeded at this date in order to obtain the greatest diversity of plant types (3).

Seed lots were sown in rows 18 inches apart in a randomized block design. Plants were blocked and thinned to one every 12 to 14 inches in the row approximately 1 month after seeding. About 185 alfalfa plants and 145 clover plants were established each year for each sub-seed lot. The foliage of the plants was sprayed frequently to control insects.

[1]Contribution from the Department of Agronomy, Wisconsin Agricultural Experiment Station, Madison, Wis. Published with approval of the Director, Wisconsin Agricultural Experiment Station.

The alfalfa plants were cut back uniformly to the crown on about September 9 of the seedling year. Without disturbing the natural direction of stem growth, the height of the terminal bud above the soil surface on the tallest stem of each alfalfa plant was measured in inches during early October prior to frost. The alfalfa plant height data obtained each fall were separated into four groups: *short*, *medium*, *tall*, and *extra tall*. In 1958, plants categorized as *short* were from 0 to 4.9 inches tall; *medium* from 5.0 to 6.9; *tall* from 7.0 to 8.9; and *extra tall* 9 inches and taller, respectively. The range of heights for each group was increased 2 inches in 1959 so that approximately the same percentages occurred in the Vernal variety during each year. The percentage of alfalfa plants in each height group was determined, and the percentage transformed to $\sqrt{\%}$ for statistical analysis. Using this procedure, highly significant correlations have been obtained (4) between per cent winter injury during the seedling winter and the average height of plants (0.94), per cent of extra tall plants (0.95), and per cent of short plants (–0.81).

The red clover plants were allowed to grow without cutting during the seedling year. The plants were classified in early October on the basis of growth habit and flowering into five plant types, in the same manner as described previously (3). Two of the plant types were non-flowering and three were flowering types. The percentage of plants in each plant type was determined and transformed to $\sqrt{\%}$ for statistical analysis. This procedure was used because red clover plants that flower during the seedling year winterkill more readily than non-flowering plants (1, 3, 5).

The topgrowth of the legumes was removed in late November to prevent differential snow accumulation over winter. Estimates of winter damage were made the following spring after the plants had begun growth. Each alfalfa plant was rated on a scale of 100 per cent for a dead plant to 0 per cent for a vigorous and uninjured plant. Red clover plants were recorded only as *dead* or *alive*.

TABLE 1. — GROWTH CHARACTERISTICS IN THE FALL OF 1958 OF SEEDLING PLANTS DERIVED FROM VERNAL ALFALFA SEED LOTS AND THE HARD SEEDS IN THEM, AND PERCENTAGE OF WINTER INJURY DURING SEEDLING YEAR

Type of seed[1]	Av. height of all plants, inches	Per cent plants in height groups				Per cent winter injury
		Short	Medium	Tall	Extra tall	
Original	5.63	42	27	18	13	15
Hard	5.32	46	27	18	9	14
Sig.[2]	*	n.s.	n.s.	n.s.	n.s.	n.s.

[1]Seed from 5 different seed lots with five replications of each
[2]Statistical significance, *significant at 5% level

TABLE 2. — GROWTH CHARACTERISTICS IN THE FALL OF 1959 OF SEEDLING PLANTS DERIVED FROM VERNAL, RANGER, AND BUFFALO ALFALFA SEED LOTS AND THE HARD SEEDS IN THEM, AND PERCENTAGE OF WINTER INJURY DURING SEEDLING YEAR

Type of seed[1]	Av. height of all plants, inches	Per cent plants in height groups				Per cent winter injury
		Short	Medium	Tall	Extra tall	
			Vernal (six seed lots)			
Original	7.20	44	30	18	8	43
Hard seed	6.83	50	31	13	6	40
Sig.[2]	**	**	n.s.	**	n.s.	n.s.
			Ranger (two seed lots)			
Original	9.58	12	24	35	29	68
Hard seed	9.31	10	32	31	27	68
Sig.[2]	n.s.	n.s.	*	n.s.	n.s.	n.s.
			Buffalo (one seed lot)			
Original	10.47	8	13	32	47	85
Hard seed	10.47	6	22	27	45	87
Sig.[2]	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.

[1]Ten replications of each
[2]Statistical significance, * significant at 5% level, **significant at 1% level

RESULTS

Alfalfa Hard Seed

The average performance of alfalfa plants derived from the original seed and from the hard seed are shown in Table 1 for 1958 and in Table 2 for 1959. Analyses of the data of both years indicated that differences were consistent from seed lot to seed lot within each of the varieties. With Ranger and Buffalo, where only three seed lots were studied, the fall growth habit and winter injury of plants derived from hard seed and from the original seed were not significantly different, except for the per cent of medium height plants of Ranger. Several seed lots of Vernal were studied. The mean height of plants from hard seed was significantly shorter than for the plants from the original seed. The populations from hard seed had a higher percentage of short-growing plants and a lower percentage of tall-growing plants than the populations from the original seed. However, the difference in per cent of short plants was significant in 1959 but not in 1958. Also, the difference in per cent of extra tall plants was not significant in either year, and the difference in per cent of tall plants was significant only in 1959. Winter injury of the plants derived from hard seed and from the original seed was not significantly different for any of the alfalfa varieties.

Red Clover Hard Seed

Medium red clover hard seeds were studied only in 1959. The average performance of the red clover plants derived from the original seed and from the hard seed showed almost identical percentages of plants in each

of five plant type groups into which the populations were categorized. Also, there was no significant difference in winter killing (original 37 per cent and hard seed 42 per cent) during the first winter.

Size of Seed

No significant differences in performance were found in 1958 or 1959 among the plants of Vernal alfalfa derived from seed fractions obtained during the cleaning of a seed lot (original seed lot, large seed, small seed, and the final seed lot). The populations were alike in mean plant height, percentage of short, medium, tall, and extra tall plants, and in winter injury, with one exception. In 1959, the plants derived from large seed had a significantly higher mean plant height than the plants derived from the other seed fractions.

CONCLUSIONS

Plant populations of alfalfa and medium red clover showed no marked differences in fall growth habit or in winter survival between populations derived from entire seed lots and from the hard seeds in them. However, the fall growth of Vernal alfalfa plants derived from hard seeds was significantly shorter in plant height than that of plants derived from the entire seed lots. No important differences were observed in fall growth habit among plant populations derived from large or small seeds of Vernal alfalfa.

Hard seeds apparently contribute plants to the population that perform similarly to those from the other seeds, and do not provide plants of higher winter-hardiness as has been suggested (2). Hard seed or seed size are not likely to be useful in screening for yield or hardiness.

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January 19, 1961

NOTE ON A GREENHOUSE TECHNIQUE FOR CROSSING LEGUMES BY HONEY BEES[1]

Honey bees have often been used for legume pollination in greenhouses. The mechanization of this procedure, however, is far from standardized. Probably the most common method is the isolation of a section of a greenhouse or the use of a growth room as a pollination chamber (3). The method described below offers many advantages over previous methods. It is highly flexible, efficient and requires a minimum of labour, space and bee management.

MATERIALS AND METHODS

The essential feature of this method is the enclosure of an individual greenhouse bench with an insect cage as shown in Figure 1. The greenhouse bench in this illustration was 3 x 16 feet and 2 feet high. The bench top consisted of heavy galvanized expanded wire mesh. This was covered with heavy wrapping paper over which a sheet of 6-millimetre polyethylene was placed to provide for sub-irrigation of the potted plants. About 1½ inches of peat moss over the polyethylene protected the plastic from damage by plant pots and prevented the bees from drowning.

The frame for the insect cage was made from Dexion and was bolted to the greenhouse bench to which it conformed in size (3 x 16 feet). It was 5 feet in height to accommodate sweetclover, the tallest legume in the

[1]Contribution No. 91, Canada Department of Agriculture Research Station, Saskatoon, Sask.

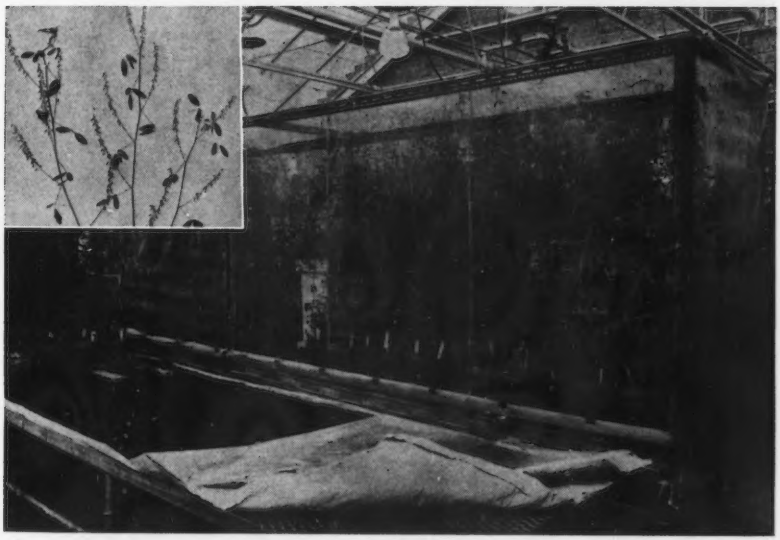

FIGURE 1. Insect cage used for cross pollination of legumes by honey bees. Bench in foreground with platform prepared with wrapping paper, polyethylene sheet and peat moss in position. Inset shows seed set on *Melilotus officinalis* from cage.

breeding program. The insect cage was made from 20 x 20 Lumite screen. Heavy duty zippers were placed at each corner as well as vertical zippers on each side to facilitate handling of plant material and bees. A draw-string at the bottom of the cage kept the base of the cage flush against the greenhouse bench. It was, however, necessary to supplement the draw-strings with foldback paper-clips to fasten the bottom edge of the cage securely to the greenhouse bench. The cost of this cage, including the Dexion frame, was approximately $60.00. The incandescent lights were located above the cage so that the bees were protected from contact with the hot light bulbs (Figure 1).

Honey bees were provided in a 5-frame nucleus (Figure 2) containing a pound of bees, a laying queen and at least 10 pounds of honey and pollen. Although queenless nuclei may be used (3), they are not as efficient and are likely to require additional bees and brood over extended periods of utilization. For short-term use, a 2-frame observation hive (Figure 2), the dimensions of which are 20 x 20 x 2 inches, with glass sides and a broad base, may be preferred as the condition of the hive with regard to bees, brood, and stores can be observed without manipulation. Bees remaining after removal of the hive can be quickly exterminated by use of a DDT or other insecticide smoke generator. Cages should be washed before further use as a precaution against insecticide residues and spread of bee diseases.

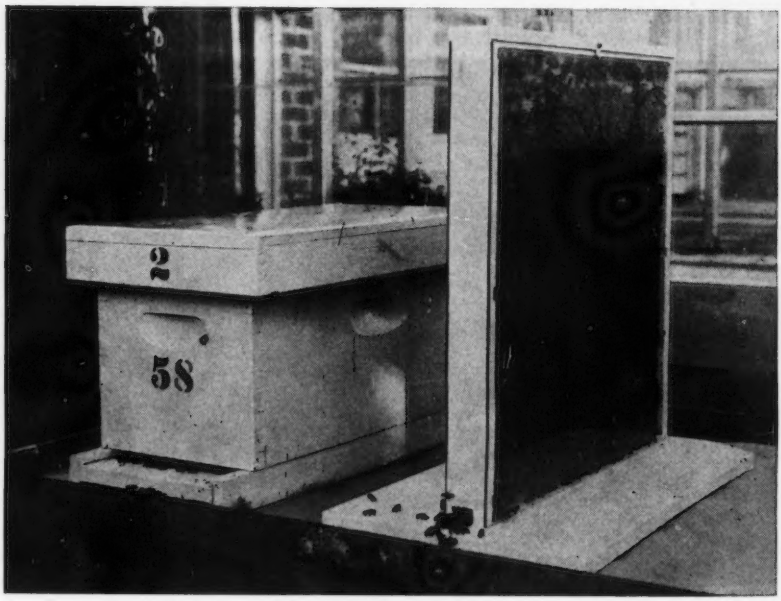

FIGURE 2. Types of hives used for greenhouse pollination of legumes. *Left* — 5-frame nucleus. *Right* — 2-frame observation hive.

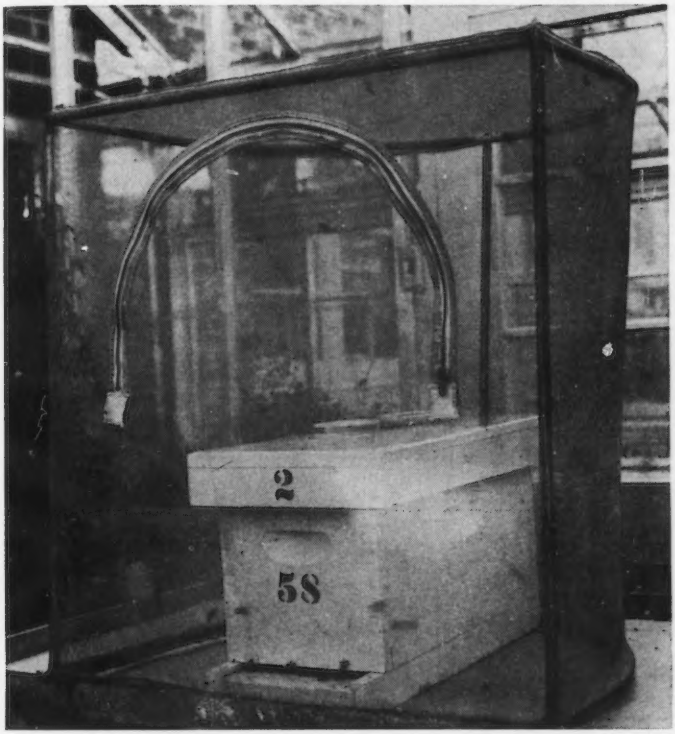

FIGURE 3. Holding cage for colony when not in use.

When not in use, nuclei may be maintained for long periods in a small screen enclosure (Figure 3) if provision is made for water and stores. Finely-ground pollen and sugar syrup given separately outside the hive are excellent stimulants for brood rearing and for "conditioning" of the bees for future flower visitation. A colony equipped with a pollen trap (4) during the summer may provide as much as 15 pounds of pollen for use in greenhouse feeding.

RESULTS AND DISCUSSION

The use of this system of intercrossing by honey bees in greenhouse cages can be illustrated by the sweetclover breeding program at Saskatoon. During the winter of 1960-61, three separate backcrossing programs were carried out concurrently in the same greenhouse section using separate cages. Each backcross involved the intercrossing of approximately 300 plants. In each cage, low coumarin and high coumarin plants were placed in checkerboard fashion to ensure maximum intercrossing. Hybrid seed was distinguished from selfed seed by the low coumarin recessive genetic marker (2). The subsequent intercrossing of known F_1 plants by honey

bees facilitated segregation to recover the low coumarin backcross derivatives. The use of honey bees for intercrossing F_1 not only obviated the tedious task of selfing by hand, but it also precluded an inherent selection for self-fertility which occurs in selfing.

Improvement of cross-pollinated species by backcross breeding involves sizable populations to assure that the sample of gametes taken from the recurrent parent represents the gene frequencies characteristic of that variety (1). Similarly, the production of synthetics involves large numbers of plants. The use of honey bees for intercrossing as described above expedites handling these large populations.

The seed set in these cages was excellent (Figure 1); ½ to 1 pound of seed was produced in each cage. This reflects the feasibility of producing synthetic seed in the greenhouse during the winter months.

The apparent specificity of the individual honey bees allows unrelated species of plants to be included in the same cage. The high population of bees in such a small area ensures crossing of plants even with low honey bee preference. Where backcrossing or hybridization is carried out on a small scale, the substitution of honey bees for hand pollination on emasculated plants may be considered.

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March 13, 1961

NOTE ON RUSSELL OATS

Russell oats (C.A.N. 844, Sel. Ott. 5055-46) was developed through the co-ordinated efforts of the Eastern Cerealists Committee and the Ontario Project Group, and was licensed on March 22, 1960. This variety is similar to Garry in many agronomic characteristics and in disease resistance. Its seed quality is good, having a large kernel of high bushel weight and low hull percentage. It is more tolerant to Septoria leaf blotch and black stem than Garry and other currently available oat varieties. It appears to be particularly well adapted to Ontario.

ORIGIN AND BREEDING METHODS

The variety originated as a selection from a backcross made at the Central Experimental Farm, Ottawa, Ontario. The first cross, (Garry-Mutica Ukraine) F_8 x Abegweit was made in 1950 and the F_1 was backcrossed to Abegweit during the winter of 1950-51. Five generations of single plant selections were grown in disease nurseries in the greenhouse and field from 1951 to 1954 inclusive. Selected lines were first entered in replicated yield tests in 1955. Two sister strains, 5055-13 and 5055-46, were advanced to the Eastern Co-operative Tests in 1957 and both strains were among the top yielders during the next 3 years in these tests[1]. Strain 5055-46 was entered in the Ontario Regional Tests in 1959. It was preferred to 5055-13 because of its earlier maturity and shorter straw. In 18 Co-operative Yield Tests conducted over a 3-year period on Ontario Stations, Russell outyielded the top recommended varieties by 5.6 bushels per acre. On the basis of its performance in Co-operative and Regional Tests in Ontario (Table 1), recommendations for application for license were made by the Ontario and Quebec Project Groups. Although generally performing well in yield tests across Eastern Canada, Russell is

[1]Yield test data are recorded in the 1957-1959 Annual Reports of the Eastern Canada Co-operative Oat Tests, Cereal Crops Division, Central Experimental Farm, Ottawa, and the 1959 Progress Report Regional Tests with Oats and Barley in Ontario, Ontario Agricultural College, Guelph, Ont.

TABLE I. — SUMMARY OF DATA FROM EASTERN CO-OPERATIVE OAT TESTS, 3-YEAR AVERAGES — 1957-1959

Variety	Yield per acre		Maturity	Length of straw	Lodging resistance	Weight per bushel	1000-kernel weight	Hull content
	Ontario, 6 stations	Eastern Can., 15 stations						
	bu.	bu.	days	in.	1-9	lb.	gm.	%
Ajax	84.9	87.3	96	43.2	3.4	35.4	27.5	27.7
Garry	87.5	87.2	99	42.3	3.0	35.9	29.4	28.0
Glen	87.8	90.9	96	42.0	3.3	34.7	34.2	27.0
Shield	80.0	80.4	93	38.4	2.6	35.6	28.6	24.7
Victory	83.2	83.7	103	45.0	3.8	35.6	30.0	28.0
Fundy	86.4	88.6	94	44.0	3.2	35.1	30.3	27.2
M.C. 6846	81.9	83.2	93	40.0	2.6	34.5	29.2	25.1
Russell	92.9	90.8	100	41.7	3.2	36.0	33.6	25.3

particularly well adapted in Ontario. Seed was distributed in 2-bushel lots to 40 Elite stock seed growers selected by the Ontario Foundation Seed Committee in the spring of 1960.

VARIETAL CHARACTERISTICS

Plant Type— Russell is a medium short, strong strawed variety with equilateral erect panicle branches and glabrous nodes. It matures about 1 day later than Garry.

Disease Reaction—It has resistance to loose and covered smut, Victoria blight, and has resistance to crown and stem rust similar to Garry. It is more tolerant to Septoria leaf blotch and black stem than other currently available varieties of oats. It is moderately susceptible to leaf spot caused by *Helminthosporium avenae*.

Grain Characters—The seed is creamy white in colour; basal hairs are absent to trace; awns are scarce and the rachilla is medium in length and glabrous. The kernel and bushel weights are high and the hull percentage is low. Fatuoids are very scarce in the present seed stocks.

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March 27, 1961

NOTE ON WILD OAT CONTROL BY DELAYED SEEDING OF WHEAT

Barley is generally recommended in a delayed seeding program for wild oats control (1). Results of experiments conducted on a heavily infested clay soil at Kyle, Saskatchewan, indicate that wheat, also, can be used successfully in such a program in areas where the growing season is normally long enough for wheat to mature following delayed seeding.

Six tests of early and delayed seeding of wheat were conducted during the period from 1954 to 1960. The early dates used in these tests corresponded closely with the normal early seeding dates in this area. All treatments were seeded with a disker, without prior cultivation and were rod-weeded 4 to 8 days after seeding. There was a greater wild oat infestation, in all years, in the early than in the late seeded wheat. Figure 1 shows the control obtained in 1957. Yield data, as presented in Table 1, were determined by using field-scale equipment or by square-yard samples. A swath was cut through the centre of the plot with a self-propelled swather or combine, and the grain weighed in a weight-recording attachment on the combine. Delayed seedings yielded more than early seedings. These increases varied from year to year and resulted in a 6-year average increase of 7.5 bushels per acre. Despite the fact that there were some variations in the early and late dates from year to year, there seems little doubt that most of the variations in yield were due to differences in wild oat infestations.

FIGURE 1. A heavy infestation of wild oats in early seeded wheat (*right*) compared to late seeded wheat (*left*). (Kyle, Saskatchewan, August, 1957).

TABLE 1.—THE EFFECT OF DATES OF SEEDING ON YIELD OF WHEAT

Year	Dates of seeding		Yield—bu./a.c.	
	Early	Delayed	Early	Delayed
1954	May 10	May 25	15.0	18.0
1955	May 12	May 25	27.0	30.5
1956	May 5	May 25	26.0	35.0
1957	May 1	May 23	9.0	22.0
1959	April 27	May 20	19.0	33.2
1960	May 5	May 20	27.4	29.9

TABLE 2.—THE EFFECT OF DATES OF SEEDING ON WILD OAT POPULATIONS

Seeding treatment	Date 1957	Wild oat plants per sq. yd. — 1957	Soil populations of wild oats bu./ac.	
			Fall — 1955	Fall — 1957
Early barley	May 1	38.5	7.3	17.1
Late barley	May 23	1.5	8.7	5.6
Early wheat	May 1	41.0	6.2	22.0
Late wheat	May 23	3.5	8.8	5.4

It is of interest to note that the largest increases in yield resulted when the interval between early and late seeding was approximately 3 weeks. Observations of the tests at harvest time revealed that wheat seeded as late as May 25 ripened during the normal harvest period.

The 1957 test included both wheat and barley in order to compare the relative efficiency of these crops in controlling wild oats. The degree of wild oat control was determined from soil populations and plant counts of wild oats. The average population of wild oats in the soil was obtained from 48 soil samples per treatment, each 4 inches in diameter and 5 inches deep. These data were obtained in the fall of 1955 and also in the fall of 1957, and are expressed on a bushel-per-acre basis (Table 2). The average number of wild oat plants per square yard was obtained from 48 samples per seeding treatment (Table 2). These data show on the average that wheat and barley were equally effective in controlling wild oats at the delayed seeding date used in this test.

The results of these tests indicate that delayed seeding of wheat is practical for wild oat control in southwestern Saskatchewan.

REFERENCE

1. Cultural control of wild oats. Bull. 1044, Can. Dept. of Agriculture, Ottawa. 1959.

—N. A. KORVEN,
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November 21, 1960

NOTE ON HOLDER-LIFTER TONGS FOR HARVESTING FORAGE ROWS

As a part of the plant breeding program at the Ontario Agricultural College, Guelph, Ontario, a very large number of birdsfoot trefoil progeny rows require harvesting in the seedling year. Because of the succulent nature of the seedlings they bend away from the mower knife and difficulty has been experienced in harvesting these rows quickly, easily, cleanly and efficiently. The sickle-bar mower, the suction mower, the scythe and hand clippers were all tried and were found to be ineffective, laborious or unacceptable in some other important attribute. A departure from orthodox methods seemed to be called for.

A solution to the problem was supplied by H. Sivers, a member of the legume breeding section at the College. Two straight boards, of either wood or of aluminum, of the required length and about 3 inches wide, were bolted to a metal framework which served both as a vice for holding and a handle for lifting. The accompanying photograph shows the design. To operate the tongs one of the boards was placed on one side of the row to be harvested, its mate on the other. If the row was upright the two boards were brought together by means of the metal handles and the tongs lifted just high enough to permit the bar of the mower to pass beneath and cut the forage. If the forage was semi-prostrate, as was frequently the case with Empire birdsfoot trefoil, a slight amount of manipulation enabled the operator to grasp this type of row with the tongs and raise it to an erect position where cutting became possible.

When the mower had done its work, the grip on the cut stems was maintained and the forage, still held between the two boards, was lifted from the row and carried to a receiving container.

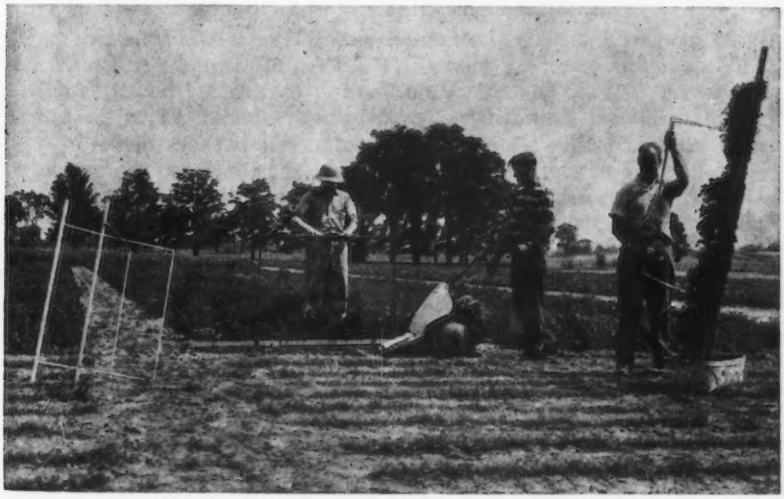

FIGURE 1

The implement proved equally effective for trefoil rows 2 months old or 2 years old. It was light to handle, economical to construct, amenable to modification in length and made possible a cutting operation that was rapid, uniform and complete.

ACKNOWLEDGEMENT

It is a pleasure to acknowledge our indebtedness to the technical staff of the Ontario Agricultural College by whom the equipment was built.

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May 5, 1961

NOTE ON CHEMICAL CHANGES OCCURRING IN FREEZE-DRIED AND FRESH-FROZEN WHEAT LEAVES DURING STORAGE

In recent years, the technique of freeze-drying has become a popular method of preservation of fresh tissues for later chemical analysis. The convenience of fresh-frozen and of freeze-dried wheat leaves as basal media for insect nutrition studies has been suggested by McGinnis and Kasting (3). Such material should remain essentially unaltered in chemical composition for extended periods for maximum benefit to be derived from it. That freeze-drying is not without its pitfalls has been recognized by relatively few workers (1, 2, 6). Laidlaw and Wylam (2) have observed that rapidly freeze-dried grasses analysed immediately after drying contain almost the same quantities of ethanol-soluble carbohydrates as do fresh plants. This carbohydrate content, however, changed considerably and erratically on storage in sealed jars at either 0°C. or room temperature. Variations of more than 100 per cent from the original amounts were noted in some cases. These writers also mention that A. R. Kemble obtained similar results in studies of "water-soluble nitrogen": storage of freeze-dried grasses for 3 months resulted in an increase in water-soluble nitrogen to almost twice the original amount. It is thus apparent that the freeze-drying of plant tissues does not inactivate all of the enzymes originally present and that at least some of these enzymes can function in the presence of the residual 5-10 per cent (2, 4) of moisture in the tissues. In this regard, Roberts[1] has observed appreciable invertase activity at –20°C. in enzyme preparations from wheat leaves.

The extent to which the chemical composition of fresh-frozen and freeze-dried wheat leaves varies with storage time and conditions was investigated. This note presents the results of these studies.

The plants[2] used were *Triticum aestivum* L. emend. Thell. var. *vulgare* cv. Thatcher; Chinese Spring; S-615; Rescue; Rescue monosomics V, IX, and XVIII; and the derived normals of V, IX, and XVIII. Freeze-dried leaf meal[3] of all varieties was prepared as described elsewhere (3) and was stored in covered jars at room temperature. All plants used were grown to the two-leaf stage in nutrient solution either in a greenhouse or in a growth cabinet. Leaves to be fresh-frozen were placed in polyethylene bags and stored in a controlled temperature room at –20°C. Control plants, immediately after harvest, were frozen in a mixture of dry ice-80 per cent aqueous ethanol and the ethanol was then heated to boiling and refluxed for 15 minutes. Two further extractions with boiling 80 per cent ethanol were made. The freeze-dried and fresh-frozen leaves were extracted similarly with the omission of the ethanol-dry ice treatment. After the pigments had been extracted into light petroleum ether, the ethanol solutions were further fractionated by ion-exchange on columns of Amberlite IR-45 and IR-120 and all fractions were examined by ascending two-dimensional paper

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[1]Roberts, D. W. A. *Personal communication.* 1959.
[2]Seeds for these plants were obtained through the courtesy of R. I. Larson, of this Station.
[3]Obtained through the courtesy of R. Kasting and A. J. McGinnis, of this Station.

TABLE 1.—THE EFFECTS OF STORAGE TIME AT ROOM TEMPERATURE ON THE CONTENT OF CERTAIN COMPOUNDS IN FREEZE-DRIED WHEAT SHOOTS, VAR. RESCUE MONOSOMIC V (R-V) AND VAR. RESCUE DERIVED NORMAL OF MONOSOMIC V (R-V-N)

Time after freeze-drying, months	Compounds, per cent dry weight							
	Sucrose		Glucose		Fructose		Free amino acids	
	R-V	R-V-N	R-V	R-V-N	R-V	R-V-N	R-V	R-V-N
0	5.6	6.3	0.7	0.8	1.0	1.4	0.2	0.4
0.5	5.2	6.6	0.5	0.8	1.1	1.5	0.2	0.5
1	5.7	5.8	0.9	0.7	1.4	1.2	0.3	0.7
3	5.4	4.1	0.4	0.4	1.9	1.6	0.4	0.9
6	5.6	1.7	0.4	1.5	0.8	2.0	0.6	1.1
12	5.0	0.3	0.8	3.3	1.3	1.1	0.7	1.3

chromatography using solvents and sprays recommended by Smith (5) for sugars and amino acids. The error associated with these determinations did not exceed ± 10 per cent.

Storage of freeze-dried wheat leaves in the presence of air at room temperature resulted in changes similar to those observed by Laidlaw and Wylam (2). The changes in content of sugars and amino acids were not large after 2 weeks of storage but became noticeable after 1 month and continued up to 1 year when the study was discontinued. Extensive and erratic changes in the content of sugars and amino acids were noted in all varieties. Typical data for two of the varieties studied are presented in Table 1. In many varieties the sucrose content fell to less than 5 per cent of the original (e.g., R-V-N, Table 1), while in others, the sucrose content remained unchanged but the glucose or fructose content increased or decreased in apparently random fashion (e.g., R-V, Table 1). In all varieties, the free amino acid content generally almost doubled during the first 3 months. The distribution of the amino acids was also slightly altered in these leaves: the most commonly observed change was a decrease in asparagine content.

The storage of fresh-frozen wheat leaves at −20°C. resulted in changes similar to those observed with the freeze-dried material. The most significant observation was that, as in the freeze-dried leaves, the changes in content of these compounds were very erratic; no straightforward relationship with time could be noted.

The changes occurring in the chemical composition of leaves stored at −20°C. are not unexpected in view of the possibility that the eutectic point of certain parts of a cell may be well below −20°C. (4). If such a system prevails, the enzymes present in the liquid phase will be active at reduced rates. The changes in the freeze-dried leaves are not surprising in view of the 5-10 per cent of water remaining in this material. Further, air in the storage containers may allow oxidation reactions to occur. Although the effects of microbiological contamination on the chemical con-

stitution of the freeze-dried meals were not assessed, such contamination may be an important factor in long-term storage of this material.

When water is added to these meals (as when they are used for insect diets), activation of many of the preserved enzymes will result and the chemical constitution of the meal (and the diet) will change continually as long as any enzymes remain active. Such changes could be very important when diet supplementation studies are being conducted. The utility and convenience of freeze-dried plant tissues as basal media for animal nutrition studies is not disputed; the essentiality of recognizing the possible alterations in chemical constitution which may occur in such media is emphasized.

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